World Journal of Fish and Marine Sciences 7 (4): 268-277, 2015

ISSN 2078-4589

© IDOSI Publications, 2015

DOI: 10.5829/idosi.wjfms.2015.7.4.95119

Quality Assessment of Fresh and Dried Puffer Fish (*Lagocephalus lunaris*) Obtained from Tuticorin, South East Coast of India

K. Immaculate Jeya Santa, Saritha. K., Hermina Giftson and Jamila Patterson

Suganthi Devadason Marine Research Institute, 44-Beach Road, Tuticorin-628001, Tamil Nadu, India

Abstract: The present study was conducted on fresh and dried fish of low value puffer fish (*Lagocephalus lunaris*) used in local export, to determine proximate and quality characteristics. Samples were collected from puffer fish processing company in both fresh and dried form. The reason for taking the puffer fish for this study is, nowadays, small scale exporters are exporting muscle of fresh and dried puffer fish, after removing skin and internal organs. But they were not worried about the quality status and they only export it in the form of chilling not in frozen forms. This study gave awareness to the exporters about the quality characteristics of puffer fish product and also improves the processing of the fishes. The proximate composition of the puffer fish in fresh and dried form had a good nutritive value but quality wise it is not attaining the standard limit it was poor based on both biochemical and microbial. It may be due the lack of knowledge about the hygienic processing method, immediate icing, or due to removal of skin manually that are more susceptible to pathogenic contaminations. So improvement in the processing procedures save the valuable protein resources and improve the foreign exchange earnings.

Key words: Fresh and Dried Puffer Fish • Lagocephalus lunaris • Export Market • Quality Assessment

INTRODUCTION

Fish is one of the most important sources of animal protein available in the tropics and has been widely accepted as a good source of protein and other elements for the maintenance of healthy body [1]. Fish protein contains all the essential amino acids and is considered to be a complete protein [2]. Fishes have easily digestible nature of proteins and are important source of essential minerals as in very fresh condition. Besides, the dried fishes are also rich in other nutritional components [3]. Laureti [4] established that dried fishes often are an alternative to fresh fishes in many places. In India significant amount of dried fishes (approximately 622 mt) were exported and earned of foreign currency [5]. However protein composition varies with different species and within the same species and with the mode of utilization. Fish is currently being used as a good tool for food therapy and source of therapeutic substances for the treatment of coronary diseases, auto-immune diseases, anemia and protein energy malnutrition. Fish is the major source of animal protein in the diet of the people of Tamil Nadu contributing 58% of the total animal protein supply.

A sizeable quantity of fish is used as fresh and some are preserved by salting and sun drying. It contributes 3.74% of the gross domestic product (GDP) and 4.04% of the foreign earnings. Tamil Nadu earns a good quantity of foreign exchange by exporting fresh and dried fish every year. In previous days for this export they prefer only economically valuable seafood's like prawns, crabs, cephalopods and fin fishes like Istiophorus platypterus, Carangoids malabaricus, Scomberoides lvsan, Lethrinus rubrioperculatus, Auxis thazard, Leiognathus dussumieri and Sphyraena acutipinnis [5]. Because of the high cost problem, small scale exporters cannot buy those things for export and they prefer economically low value fishes for export. In Tuticorin coast normally high amount of puffer fishes coming along with the by-catches this would be treated as trash, which are dumped with other uneconomical fishes. These fishes are weighing of about nearly 250g each. In past the fishes are considered as poisonous and so many disease outbreaks causes by intake of puffer fish [6]. The person affected usually experience vomiting, dizziness and weakness and potentially death [7]. Recent studies have revealed that the liver of puffer fish

has a specific tetrodotoxin (TTX)-uptake mechanism and TTX introduced into the puffer fish body is first absorbed in the liver and then transferred to the skin through the circulatory system. Eswar et al., [8] and Li et al., [9] reported puffer fish can act as a good source of nutritional and economic value when it is carefully handled and properly cooked. These fish contain 'tetrodotoxin' in the skin and visceral organs like gonads and intestine. Tetrodotoxin is named after the order of fish from which it is commonly associated, the Tetrodontiformes fish contain highly toxic, this fish are eaten by some Japanese. If cleaned properly, the puffer fish flesh is fit for human consumption and considered daintiness [10]. So now a day instead of wasting them, these puffer fish can also be used as food to meet the nutritional requirements of increasing population by removing the skin and visceral organs which are suspected to contain the toxin. Small scale exporters in Tuticorin prefer this fishes to export only the flesh in the form of fresh and dried. Good quality raw material supply is essential for both domestic and value added product development for export market [11].

The quality of fishes can be estimated by sensory tests, microbial methods or by chemical methods such as measuring volatile compounds, lipid oxidation and determination of ATP breakdown products and the formation of biogenic amines [12]. The increasing demand in the commodity as well as the need to protect the consumers against food-borne diseases have made regulatory authorities such as the United States Food and Drug Administration and the European Union stipulate stringent guidelines with respect to the quality of processed fish items traded by exporting countries. Hence, many developing countries attempt to set up standards for their fishery product in order to comply with requirement of international trade and markets [13]. But in the case of small scale traders export the product to the local market without meeting the quality standards. This study will be helpful in producing a quality product as well as safe product for domestic consumption as well as for export. Now-a-days consumer wants to know and ensure the nutritional value of the products, what they are eating. At the first step, the result of the present investigation is expected to provide a clear idea on the quality and safety of the fresh Lagocephalus lunaris fish under present study. The nutritive value of the puffer fish is already established [8]. A better knowledge on quality and safety of sundried and fresh puffer fish Lagocephalus lunaris is important because a reasonable quantity of fresh and dried fish is exported to international markets every year. To continue

export of this fishery product the quality and safety of the product should be assured. So in the present study was undertaken to know the quality of low value puffer fish preferred to export both in fresh and dried form

MATERIALS AND METHODS

Sample Collection and Storage: The fresh puffer fish were purchased from local export company. In this company puffer fish (Lagocephalus lunaris) were purchased from Tuticorin fishing harbor and other minor landing centers of Tuticorin. The samples were transported to the processing center with sprinkling the ice on the top of the fish box. In the processing centers, the skin and visceral organs like gonads and intestines were removed and only the muscles were used as an edible portion. For the experimental studies the puffer fish muscle samples were transported immediately to the laboratory for the analysis.

The sundried fish of *Lagocephalus lunaris* were also purchased from the same processing center of Tuticorin. The dried fish samples were packed tightly in polythene bags and stored at -20°C until further analysis for subsequent studies. The samples were subjected to laboratory analysis within 2 weeks of purchase.

Biochemical Composition Analysis: Organoleptic analysis: The organoleptic characteristics of fresh and dried fish products observations were done according to Geetha *et al.* [14].

Biochemical analysis: Analytical methods were applied for the determination of biochemical composition of the processed fish products on experimental basis. The analytical methods are given below: The moisture content of all the samples was analyzed by drying the samples in a hot air oven. The protein content of the samples was estimated by Lowry's method [15] and lipid by using gravimetric method [16], the ash content was measured by the method of Clucas and Ward [17] using Muffle furnace. Carbohydrate content was determined by the method of Hedge and Hofreiter [18] using Anthron and hydrochloric acid.

Mineral Contents: The minerals in the homogenous fish powder were brought into solution by wet digestion using the method of Harris [19]. Potassium and sodium were determined by Allen's method using Perkin-Elmer atomic absorption Spectrophotometer [20]; phosphorous by Bausch-Lomb spectronic 20 [20]; zinc, calcium, iron, magnesium and copper by using Perkin-Elmer atomic absorption Spectrophotometer [21].

Quality Analysis: Chemical changes were studied by determining the TMA-N and TVB-N using Conway modified micro-diffusion technique [22]. Free fatty acid content of the samples was estimated by improved titrimetric method of Ke *et al.*, [23] as described by Takagi *et al.* [24]. Peroxide value was estimated by the method described by Ozogul *et al.* [25]. Peroxides value of the samples was expressed as milimoles of oxygen per kilogram of fat. pH analysis was done by the method of Goulas and Kontaminas [26] using HANNA pH213 microprocessor pH meter.

Microbiological Quality Analysis: The microbiological characteristics such as total plate count (TPC) were enumerated by using plate count agar and total fungal count (TFC) was enumerated using potato dextrose agar (APHA, 1992). The pathogenic bacteria like *Coliforms, Streptococci, Staphylococci, Escherichia coli, Salmonella* and *Vibrio* were enumerated by the method [27]. All determinations were done in triplicate and the mean value was reported. The result was performed from following formula-No. of bacteria (CFU/g) = No of Colony × Dilution Factor

RESULTS AND DISCUSSION

Fish is a low acid food and is therefore very susceptible to the growth of 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 India, 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 [28]. Preservation process starts when it is harvested and become complete when reaches the consumer's table. Among the different fish products, fresh and dried fish is an important source of animal protein in Tamil Nadu. The exports to the European countries have certain quality norms but the small scale exporters did not worry about that [29]. The quality of fish landings in the country is generally poor and wastage is high, because icing facility was given only to economically high valued fishes. About 25-30 % of the catch landed by the boats is of poor quality, as the fish holds of these boats are not refrigerated. These boats aim at quantity rather than quality and they sell the poor quality or spoiled fish to dry fish processors at low price. Lack of knowledge regarding improved fish handling and post-harvest practices has contributed to the poor quality of fish and fishery

Table 1: Oraganoleptic characteristics of fresh and dried puffer fish

Sources	Fresh fish	Dried fish
Colour	Whitish shiny	Dull white color
Uses of chemicals	No	No
Odour	Fishy characteristic odour	Fishy odour
Texture	Firm and Flexible	Tough and springy
Infestation	No	No
Broken pieces	No	No
Overall quality	Good	Good

Table 2: Biochemical composition of fresh and dried puffer fish

Parameters	Fresh fish	Dried fish
Moisture (%)	78	17.5
Protein (%)	18.2	43
Lipid (%)	9.0	2.34
Ash (%)	0.64	9.6
Carbohydrate (%)	7%	-
Potassium (mg/100g)	569	610
Sodium(mg/100g)	256	266
Calcium (mg/100g)	2541	2560
Magnesium (mg/100g)	98	112
Iron (mg/100g)	39	40
Zinc (mg/100g)	26	29
Copper (mg/100g)	1.8	2.0
Phosphorus (mg/100g)	14.2	19.4

products [30]. The processing such as skin removal, salting and drying was not done in the hygienic area, so the present study was carried out to create awareness and improve the quality characteristics of puffer fish.

The organoleptic observation of fresh and dried puffer fish were presented in Table 1. Little differences were observed between the colour of the fresh and dried puffer fish. Firm and flexible nature of texture was observed in fresh fish and tough springy nature was observed in the dried fish. There were no insects, infestation or broken pieces were found around the product. Geetha *et al.* [14] reported poor organoleptic quality was observed in fishery products based on poor quality raw material, unhygienic processing, excess drying, improper drying and handling.

Proximate composition of puffer fish products was reported in Table 2. The moisture content seems to be an exact indicator of susceptibility of a product to undergo microbial spoilage. It has been reported that moisture content was more in fresh fish than in dried samples. The natural sun drying usually takes three days for proper drying of fish and drying reduced the moisture content of the fish to 25% and if further dried to 15%, the growth of mould will cease and thereby it increases the shelf life [31]. Frazier and Westhoff [32] stated that, generally no

microbe could grow in dried products with moisture content below 15%. The obtained results of the present study revealed the moisture content of dried puffer fish were much higher than normally prepared from freshly dried products. The other reason is that the dried products were not properly packed so that they are more susceptible to microbial contaminations. In the tropical place like Tamil Nadu where relative humidity is always high and there is chance of uptake moisture from the environment. Excessive moisture uptake increases the water activity which facilitates the growth of microorganisms and reduces the nutrient and shelf life of dried products [26]. Chaijan [33] observed the moisture content level at 78.88 g in dorsal, 81.67 mg/g in ventral and 75.51 mg/kg lateral line region of fresh Pangasianodongigas. In the present study also 78% of moisture content was observed in fresh puffer fish indicates the fish was not deteriorated.

The crude proteins content in fresh puffer fish sample was 18.2% and it was increased in dried samples (43%). Li *et al.*, [9] reported protein content of farmed puffer fish was 18.44% in meat and 17.15% in testis and very low concentration in liver (3.89%). Increase of protein was due to dehydration of water molecule between the proteins causing aggregation of protein and thus results in the increase in protein content of dried fishes [34].

The lipid content in fresh sample was 9.0% and varied in dried fish samples (2.34%). Li et al. [9] reported that lipid content of farmed fish was high 63.86mg/100g in liver, 1.31 mg/100g in muscle and 1.82 mg/100g in testis. The lipid contents were lower in dried samples than the fresh fish; the variation could be the result of evaporation of moisture content with lipids. The fat content may be reduced with the evaporation of moisture content and increase during heat treatment. Lipid content varied greatly among the dried fish species, which was also reported by Stansby [35]; Kalamani and Kamasastri [36] (3.7-17.8%); Azam et al. [37] (97.7-26.13%) for other species. Shahiduzzaman et al.[38] reported that the Batashi fish (*Clupiso maatherinoides*) contains 3% lipid. Dried Rita rita contains 13.92% lipid and dry fatty fishes contain 7.10% of lipid in average [39].

The ash content varied between the fresh samples (0.64%) and the value was 9.6% in dried samples. Average ash content in the samples obtained from the study area was high as generally expected for dried products. This is perhaps due to contamination with sand and filth during drying. Clucas and Ward [17] reported that inorganic contents remain as ash after the organic matter is removed by incineration. Natural sun drying was done in open

space which allows settling of wind borne dust, insect and bird infestation. It would increase the inorganic contents in the samples and this may be the major reason for higher ash content in the dry samples.

Love [40] described that fish store most of their carbohydrate reserves in liver only as glycogen and the levels in muscle as glucose and glycogen. Fresh sample had a carbohydrate level of 7%, whereas the samples of naturally sun dried sample contain 0% of carbohydrate level. The result obtained in this investigation is more or less in agreement with the general rule formulated by Standsby [35] where there is an inverse relationship exists between the oil and moisture content of fish. The amount of carbohydrate in fish muscle is generally too small to be of any significance in the diet. The carbohydrate level of the Puffer fish were varied from 1.87 % to 1.96 % in L. lunaris and L. inermis[8]. Similar works were done by some other fish species. Nurnadia et al. [41] were recorded the carbohydrate concentration in marine pelagic fish Fringescale sardinella from west coast of Peninsular Malaysia. This Pelagic fish had 3.07 % of carbohydrate. The carbohydrate level in catfish Clarias gariepinus have been recorded in the level of 14% in fresh, 5. 48 % by raw drying method, 2.78 % by Kiln-dried method and 3.84 % by electric- dry method [42].

Concerning to the minerals, K was the most abundant macro element in fish samples and at the same time Zn accounts for the majority of the microelement in the sample. This was agreed with the results of farmed puffer fish studied by Li et al., [9]. The values of calcium, potassium, copper, zinc and iron recorded for all the fish were significantly higher than the reference values of WHO [43]. The fresh fishes of M. vollenhovenii and P. clarkii have the mineral content Ca (840mg/100g) and (1625mg/100g) and Fe levels was (119mg/100g and 35mg/100g and Mg levels 450mg/100g and 620mg/100g respectively [44]. In the present study the fresh and dried puffer fish have the mineral content Ca (2541 mg/100g) and (2560 mg/100g), K levels (569 mg/100g) and (610 mg/100g) and Na levels (256 mg/100g) and (266 mg/100g), Mg levels (98 mg/100g) and (112 mg/100g), Fe levels (39 mg/100g) and (40 mg/100g), Zn levels (26 mg/100g) and (29 mg/100g), Cu levels 1.8 mg/100g and 2.0 mg/100g, Phosphorus levels (14.2 mg/100g) and (19.4 mg/100g). The observations in dietary mineral suggest that the puffer fish samples could provide a significant proportion of calcium if consumed regularly. Ca in conjunction with P, Mg, Mn, vitamin A, C and D, Chlorine and protein are involved in the bone formation, but ca is the principal contributor. It plays important role in blood clotting in

Table 3: Quality characteristics of fresh and dried puffer fish

Parameter	Fresh fish	Dried fish
TMA-N (mg/100g)	6.8	11.53
TVB-N (mg/100g)	13.48	28.11
FFA(% of total lipid as oleic acid)	2.3	6.99
Peroxide value (millimoles of O ₂ /kg fat)	7.3	21.44
pH	7.2	6.3

muscle contraction and in certain enzymes in metabolic processes. The samples were good sources of magnesium, sodium and potassium. Magnesium is an activator of many enzyme systems and maintains the electrical potential in nerves [45]. Potassium is primarily an intracellular cation, in large part this cation is bound to protein and with sodium influences osmotic pressure and contributes to normal pH equilibrium. Potassium and sodium are widely distributed in foods with plant containing less than animal sources. When the amounts of these minerals were compared with the suggested values all the samples can be considered as good sources of these minerals if it is fresh and processed hygienically.

The chemical assessment of oxidative and hydrolytic rancidity was carried out on fresh and dried puffer fish are shown in Table 3. Total volatile bases (TVB) are a group of biogenic amines formed in non-fermented food products during storage [46]. The combined total amount of ammonia (NH₃), dimethylamine (DMA) and trimethylamine (TMA) in fish is called the total volatile base (TVB) nitrogen content of the fish and is commonly used as an estimate of spoilage and has been widely used as an index for freshness of fish [47]. The increase in the amount of TVB is parallel with the increase in TMA during spoilage. As the activity of spoilage bacteria increases after the death of a fish, a subsequent increase in the reduction of TMAO to TMA [48]. The source of DMA and TMA in fresh and processed fishery products is trimethylamine oxide (TMAO). TVB-N is present in very small quantity in fresh fish and produced mainly due to bacterial action and is mainly constituted by ammonia in the muscle produced by deamination of muscle adenylic acid and by process leading to denaturation of muscle protein. In spoiling fish, volatile bases are produced by putrefactive process and are determined as a measure of content of spoilage which the fish has undergone. Wallace [49] has pointed out that TVB-N is better index of spoilage. In the present study trimethylamine nitrogen and total volatile base nitrogen of fresh fish was 6.8 and 13.48 mg N/100g and dried fish was 11.53 and 28.11 mg N/100g respectively. TMA-N and TVB-N content of both fresh and dried fishes was found below the level suggested by different researchers for various fish and fish products. A value of 35 mg N/100g of TVB-N and 15 mg N/100g of TMA-N has been suggested border line [50]. TVB-N is mainly contributed by ammonia in the muscle produced by deamination of muscle proteins [51]. In the present study, increases of both TMA-N and TVB-N after processing into dried fish may be due to microbial activity, absorption of moisture and relative decrease in salt content. It is difficult to fix the limit of TVB-N for cured products due to variety and diversity of products and their processing procedure. In the experimental fishes also due to the processing effects little amount of volatile amine production was occurring.

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 [52]. The FFA value of fresh puffer fish was 2.3% (oleic acid percentage) and 6.99% for sun dried puffer fish. FFA values increased during the processing time. Lipid hydrolysis by itself has no nutritional significance but the accumulation of free fatty acids in fish oils in undesirable amount due to increased susceptibility to oxidation and consequent development of off flavors [53]. Bimbo [54] suggested the acceptable limit of FFA was 2-5% and in this study, the FFA value become unacceptable in fishes after salting and drying. Lipid hydrolysis occurred to a great extent at the end of the storage period, as the increase of FFA content of the samples reveals the loss of freshness [55] and the formation of FFA does not lead to the nutritional losses [56]. The results of free fatty acids indicated that salting and drying conditions accelerate lipid oxidation. A high level of FFA is a characteristic of product that have undergone both microbial and biochemical spoilage. There has also been a long debate over claims that lipid hydrolysis and associated accumulation of FFA contributes to accelerated protein denaturation in fish [57].

Oxidation rancidity is most often measured by 2-thiobarbituric acid (TBA), peroxide value (PV) or carbonyl value [58]. Peroxide value was used to determine the quality of fat and it is widely used as an indicator for the assessment of degree of primary lipid oxidation and the values expressed as millimoles or milliequivalents of active oxygen per kg of fat [33]. Connell *et al.* [59] has suggested that if PV is alone 20 millimoles of oxygen per kg of fat, then the fish may show off odour and rancid taste. In the present study lipid oxidation was observed in both fresh and dried fish, but the amount was slightly exceeding the acceptable limit only in dried puffer fish.

The high degree of unsaturation in the form of multiple double bonds in fatty acids renders fish highly susceptible to oxidative rancidity [60]. Bernardez *et al.* [61] 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 result indicates that the salting and drying conditions accelerate the lipid oxidation and this is in agreement with the results of previous workers [62 and 63] and factors that influence lipid oxidation were reported by Burlakova *et al.* [64].

pH value is a reliable indicator of the degree of freshness of spoilage. The pH in fresh condition in fish flesh is almost neutral [65]. 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 sundried salted fish was 6.3 in the present study. Because when salt is added with the fish, pH value decrease due to increase of acidic compound and after that during the storage pH value increases in the time due to increases of basic compounds. The limit of acceptability of fish products is usually 6.8 to 7.0 [66]. While the slight increases of pH in fresh fish may be due to processing of fish manually in room temperature and method of salting is responsible for decreases the pH in dried fish.

For food quality of fresh and dried fish products was analyzed by determining the bacteriological aspects are very important. Microbiological analysis also showed variation among the samples. The total bacterial count, total fungal count, E. coli, Salmonella, Vibrio, Coliforms, Staphylococci, Streptococci, were assessed in both fresh and dried puffer fish and the result a were presented in Table 4. The result of microbial analysis showed that the TPC and TFC were high in fresh fish compared to dried fish. It reveals that due to the unhygienic handling like removal of skin and visceral organs from the fresh fishes by manually and this would be responsible for the higher bacterial and fungal count. Mansur et al. [67] determined the total bacterial count of fresh and traditionally dried fishes ranged from 1.0×10⁵ and 1.0×106 cfu/g which is agreement with the present study. Taking the 10⁷cfu/g as the upper acceptable limits for fresh and frozen fish and cold smoked fish species [68], the both fresh and dried puffer fish was considered acceptable quality. In fresh kursa, 1.29×10^5 cells/g Total plate count (TPC) and 0.63 \times 10² cells/g TFC has been reported by Litlabati *et al.* [69]. Not all fungi which occur in fish are considered deleterious. Moulds are one of the important causes of

Table 4: Microbiological quality characteristics of fresh and dried fish

Parameter	Fresh fish	Dried fish
Total bacterial count (cfu/g)	3.7×10^{6}	2.1 × 10 ⁵
Total fungal count (cfu/g)	1.5×10^6	5.5×10^4
Coliforms	+	+
E. coli	+	+
Faecal streptococci	+	+
Staphylococcus	+	+
Salmonella	+	+
Vibrio	+	+

spoilage of salted dried fish products and they produce mycotoxins and they are able to grow in salt concentrations between 5 and 26% [70]. In the present study fungal count was found in both fresh and dried fish. Products deteriorate by growth of moulds if the water content is approximately 15% [71]. These observations were in close agreement to the present study. The rapid reduction in the water activity (aw < 0.75) is the most important factor in controlling fungi/mould contamination of the fishery products during storage [72]. Although, there are several reports on increases of consumers being poisoned by mycotoxins in fishery products; there is a definite risk to human health considering how fish are traditionally processed. In the present study fungal colonies were observed in the local export quality fresh and dried puffer fish having contamination. This may be due to post harvest delay, improper transportation, unhygienic handling and processing during the salting and sun drying process, contaminated working floor, salt and water. Patterson and Ranjitha [73] observed higher fungal contamination in commercial sun dried fishes of Tuticorin dry fish market.

Some dominant food borne pathogens such as E. coli, Streptococci, Staphylococci, Salmonella and Vibrio were also identified. Thus, fishery products have also been recognized as carriers of health hazards such as disease causing microorganisms Salmonella sp. and Vibrio sp., in addition to parasites, natural toxins, heavy metals and other pollutants. High microbial load in raw fishes indicates that raw fish would decompose very quickly at ambient temperature and the presence of coliforms, staphylococci and salmonella indicates the raw fish handling is not safe. Fish is thus a product that needs proper handling and processing in order to preserve nutrients and its functional components that promote good health. The heat applied during drying cause considerable reduction of microorganisms of various types. Drying by heat usually destroys all yeasts and most of the bacteria, but spores of some bacteria and molds usually survive.



Fig. 1: Puffer fish processing steps

Immaculate et al. [74] studied the microbial quality of commercially available dried fishes of India and reported the presence of microbes. The presence of higher fungal colony becomes the possible source of fungal toxins transmission among consumers. Fungal colony generally grow faster and are more resistant to high temperature and low water activity and tend to dominate spoilage in warmer climates [75]. In our present investigations microbial load in the samples from local processing centers was high due to clean and safe practices that were not followed properly. In general, fish are risky commodities which are difficult to handle and distribute. However, if fresh fish is handled properly it will rarely lose its wholesomeness. Good quality raw material supply is essential for both domestic consumption and value-added product development for export market. It needs proper research support to produce safe and quality product for export.

However, the results of the present investigation state that the biochemical composition is quite satisfactory in the fresh and dried fish species. From these results, it can be concluded that puffer fish, both fresh and dried can provide satisfactory nutrition to the nation, but the quality should be improved while during processing.

CONCLUSION

In the present study, processing of both fresh and dried puffer fish is carried out in an unhygienic condition. If modified processing is followed with maintaining the proper hygiene and sanitation, the produced products will get higher price. The quality and safety of fleshes both in fresh and dried products highly the health desirable in conscious people in the country and to achieve this improved method of hygienic practicing and maintenance of freshness should practiced be throughout the processing centers. Based on this conclusion and filed observations, this study recommended promotion of sanitation and hygiene campaign in all fresh and dried fish outlets whether local or out markets including fresh fish vendors through regular visitations and training by quality regulators.

ACKNOWLEDGEMENT

The authors are grateful to Dr. J.K. Edward Patterson, Director, Suganthi Devadason Marine Research Institute for providing facilities.

REFERENCES

- 1. Andrew, A.E., 2001. Fish quality, Fish processing Technology, University of Ilorin, Press Nigeria. pp: 7-8.
- 2. Gopakurnar, K., 1997. Tropical Fishery Product: Science publishers, Inc. P.O. Box 699, Enfield, New Hampshire 03748, U.S.A.

- 3. Basu, K.P. and K. Gupta, 2004. Biological value of protein of some species of Bengal fish by balance and growth methods. *J. Indian. Chem. Soc. Calcutta*. pp: 543-548.
- 4. Laureti, E., 1998. Fish and fishery products: World Apparent Consumption Statistics Based on Food Balance Sheets (1961-1993). FAO Fisheries Circular, 821: 3, Rome.
- Sinduja, P., K. Immaculate, J.K. Patterson and P. Jamila, 2014. Effect of gamma irradiation on the microbial quality of dried fishes. The Asia Journal of Applied Microbiology, 1(3): 26-48.
- 6. Mak, C.K. and J. Ho, 2006. A case of puffer fish poisoning. Commun. Dis. Watch, 3(23): 91.
- 7. Parvaneh, H., M. Ho Chin, N.N. Wan and A.M. Nor, 2012. Make the deadly yellow puffer fish a safe food to eat. Journal of Food, Agriculture and Environment, 10(3 and 4): 72-77.
- Kathirvel, Eswar, A., K. R. Anbarasu, K. Ramamoorthy, G. Sankar, S. Suvitha and T. Manikandarajan, 2014. Proximate composition Fatty acid analysis of Puffer fish, Lagocephalus inermis (Temminck and Schlegel, 1850) and Lagocephalus lunaris (Bloch and Schneider, 1801) from Parangipettai, Southeast coast of India International Letters of Natural Sciences, 17: 21-29.
- 9. Li, Y., L. Wang and N. Tao, 2014. Analysis and Evaluation of Nutritional Composition of Farmed Male Pufferfish (*Takifugu obscurus*) SHS Web of Conferences 6, Public Health in Rural, 7: 10-30.
- Torado, T.A., E. Sinclair and D.B. Ulyatt, 1973.
 Puffer fish (Tetrodotoxin) poisoning: Clinical record and suggested management. Medical Journal of Australia, 1: 599-602.
- Mohammad, A.M., R. Shafiqur, N.A.K. Mohammad, R. Md. Shaheed, K. Kamrunnahar and U. Shoji, 2013. Study on the quality and safety aspect of three sun dried fish. African Journal of Agricultural Research, 8(41): 5149-5155.
- Gulsun, O., K. Esmeray, O. Serhat and O. Fatih, 2009. Sensory, microbiological and chemical assessment of the freshness of red mullet (*Mullus barbatus*) and goldband goatfish (*Upeneus moluccensis*) during storage in ice. Food chemistry, 114: 505-510.
- Sulieman, H.M.A., L.O.A. Bari and M.A. Hafiz, 2012. Determination of quality and shelf life of three marine fishes (Coral trout, Greasy grouper and Red mouthed bream) based on Total Volatile Nitrogen test (TVN). Journal of life science and biomedicine, 2: 187-191.

- 14. Geetha, S., V. Govinda Rao, N. Muddula Krishna, N. Ram Sai Reddy and K. Ramesh Babu, 2014. Some aspects of biochemical and microbial analysis of sundry fish *Trichiurus lepturus* linnaeus, 1758 from the east coast off visakhapatnam. International Journal of Biological Research, 4(4): 462-465.
- 15. Lowry, O., Rose, B.H., Fart, N.J. and R.J. Randall, 1951. Protein measurement with the Folin phenol reagent. Journal of Biological Chemistry, 193: 265-275.
- Folch, J., M. Lees and S.G.H. Bloune, 1957. A simple method for their isolation and purification of total lipids from animal tissues. Biological chemistry, 266: 497-509.
- 17. Clucas, I.J. and A.R. Ward, 1996. Post Harvest Fisheries Development; A Guide to handling, preservation, processing and quality. Natural Resources institute. U.K., 5: 428.
- 18. Hedge, J.E. and B.T. Hofreiter, 1962. Determination of reducing sugars, methods in carbohydrate chemistry, *M.L Academy press*, New York, 1: 388-389.
- 19. Harris, E. 1979. Nutrition Research Techniques for Domestic and Wild Animals. Utah, USA, pp: 140.
- Allen, S.G., 1974. Chemical analysis of ecological materials. Blackwell Scientific Publications Oxford, UK Oxford, pp: 180.
- 21. AOAC, 1990. Association of official analytical chemists. Official methods of analysis of the Association of Official analytical Chemists (13th edition). Arlington, V.A., 15: 32-36.
- 22. Conway, E.J. and A. Byrne 1993. Micro-diffusion analysis of TVN, *Biochem. J.*, 27: 419-429.
- Ke, P.J., A.D. Wovewoda, L.W. Regier and A.G. Ackman, 1976. Env. Canada Fish. Marine Serb. Technol. Branch, Halifax, New Series Circular, 61: 1-6.
- Takagi, T., K. Hayashi and Y. Itabashi, 1984.
 Toxic effect of free unsaturated fatty acid in mouse assay of diarrhetic shellfish toxin by intraperitoneal injection. Bull. Japan Sci. Fish., 50: 1413-1418.
- 25. Ozogul, Y., E.B. Boga, B. Tokur and F. Ozogul, 2011. Changes in biochemical, sensory and microbiological quality indices of common Sole (*Solea solea*) from the Mediterranean Sea during ice storage. Turk. J. Fisheries Aqua. Sci., 11: 243-251.
- Goulas, A.E. and M.G. Kontominas, 2005. Effect of salting and smoking-method on the keeping quality of chub mackerel (*Scomber japonicas*): biochemical and sensory attributes. Food chemistry, 93: 511-520.
- 27. USFDA, 1995. Bacteriological analytical manual. (8 edition), AOAC International Gathers burg, USA, 401: 614-619.

- Hossain, M.B., S.N. Amin, M. Shamsuddin and M.H. Minar, 2012. Use of Aqua-chemicals in the Hatcheries and Fish Farmers of Grater Noakhali, Bangladesh, Asian J. of Animal and Vet. Ad., 8(2): 401-408.
- 29. Venugopal, V., D. Doke and P. Thomas, 1997. Thermostable water dispersions of shark meat and its application to prepare protein powder. Aqua. Food Product Technol, 6: 53-55.
- 30. FAO, 2013. Fishery and aquaculture country profiles, Sri Lanka. Food and Agriculture Organization of the United Nations, Rome, Italy.
- 31. Glucas, I.J., 1982. Present fish drying techniques in Zambia and suggested improvements. A report prepared for fisheries development project. Rome. FAO F. J. Zam (73/00/3FAO), pp: 25.
- Frazier, W.C. and D.C. Westhoff, 1978. Microorganisms important in food microbiology. In: Food Microbiology, Third Edition, McGraw-Hill Book Company. New York, pp: 539.
- 33. Chaijan, M. 2008. Review: Lipid and myoglobin oxidations in muscle foods. Songklanakarin Journal of Science and Technology, 30: 47-53.
- 34. Ninawe, A.S. and K. Rathnakumar, 2008. Fish processing technology and Product development, Impact of curing, 5: 142 (1st edition).
- 35. Stansby, E., 1962. Composition of Fish. In: *Industrial Fishery Technology*. Ed. Stansby, M. E., Reinhold Publishing Corporation, Chapman and Hall Ltd., London.
- Kalaimani, N. and P.V. Kamasstri, 1998.
 Quality characteristics of cured fish of commerce. J. Fish. Technol., 25: 54-57.
- 37. Azam, K., M.Z. Basher, M. Asaduzzaman, M.H. Hossain and M. Y Ali, 2003. Biochemical quality assessment of fourteen selected dried fishes. Univ. J. Zool. Rajshahi Univ., 22: 23-26.
- Shahiduzzaman, M., N. Banu, M.M. Hossain, M.S. Islam, M.K. Alam and M.A. Hossain, 2004. Seasonal variation in proximate composition and mineral content of *Clupiso maatherinoides*. Bang. J. Life. Sci., 16(1): 109-113.
- 39. Mollah, A.H., M.S. Rahman and M.T. Alam, 1998. Study of proximate chemical analysis of Bangladeshi freshwater fish *Rita rita* (Ham.) and seasonal variation of lipid, protein and related substances. Univ. J. Zool. Rajshahi Univ., 17: 1-6.
- 40. Love, R.M., 1980. The chemical biology of fishes. (Vol 2). Academic Press, London.

- 41. Nurnadia, A.A., A., Azrina and I. Amin, 2011. Proximate composition and energetic value of selected marine fish and shellfish from the West coast of Peninsular Malaysia. International Food Research Journal, 18: 137-148.
- 42. Ogbonnaya, C. and M. Ibrahim, 2009. Effects of drying methods on proximate compositions of catfish. World J. Agric. Sci., 5(1): 114-116.
- 43. WHO, 1974. Recommended intakes of nutrients, requirements of energy, protein, vitamins, calcium and iron for humans of all ages. Report of a joint FAO/WHO Expert group FAO, Rome, pp. 315.
- 44. Abulude, F., L. Lawal, G. Ehikhamen, W. Adesanya and S.L. Ashafa, 2006. Chemical composition and functional properties of some prawns from the coastal area of Ondo state, Nigeria. Electron. J. Environ. Agric. Food Chem., 5(1): 1235-1240.
- 45. Shils, M.E., 1997. Magnesium. In: O'Dell, B.L.; Sunde, R.A. Handbook of nutritionally essential minerals. New York: Marcel Dekker.
- 46. Horsfall, M., B.S. Kinigomaand A.I. Spiff, 2006. Evaluation of the levels of total volatile bases and trimethyleamine formed in fish stored at low temperature. Chemical society of Ethiopia, 20: 155-159.
- 47. Wu, T.H. and P.J. Bechtel, 2008. Ammonia, Dimethylamine, Trimethylamine and Trimethylamine Oxide from raw and processed fish by-products. Journal of Aquatic Food Product Technology, 17: 27-38.
- 48. Yusuf, A.M., M.I. Sharif, K.A. Ripon and O. Faruque, 2010. Post mortem variation in total volatile base nitrogen and trimethylamine nitrogen between Galda (*Macrobrachium rosenbergii*) and Bagda (*Penaeus monodon*) University Journal of Zoology, Rajshahi University, 28: 7-10.
- Wallace, H.A., 2000. Microbiological methods. In Horwitz, W. (ed.). Official Methods of Analysis of AOAC International. 17thedn. Vol. 1. AOAC International, Gaithersburg, Md., pp: 126-130.
- 50. Ghaly, A.E., D. Dave, S. Budge and M.S. Brooks, 2010. Fish spoilage mechanism and preservation techniques review. Am. J. Appl. Sci., 7(7): 859-877.
- 51. Chaijan, M., S. Benjakul, W. Visessanguan and C. Faustman, 2006. Changes of lipids in sardine (*Sardinella gibbosa*) muscle during iced storage. Food Chem., 99: 83-91.
- 52. FAO/ SIFAR, 2001. Non-Sensory Assessment of Fish quality, (FAO in partnership with support unit for International Fisheries and Aquatic Research, SIFAR, Torry Advisory, pp. 92.

- 53. Nair, P.G.V. and M. Suseela, 2000. Biochemical composition of fish and shell fish, In: CIFT-Technology advisory services, Central Institute of Fisheries Technology, Cochin., pp: 281-289.
- 54. Bimbo, A.P., 1998. Guidelines for characterizing food grade fish oil. Int. News Fats, Oils Relat. Mater. 9: 473-483.
- Ozogul, Y., G. Ozyurt, F. Ozogul, E. Kuley and S. Polat, 2005. A Freshness assessment of European eel (*Anguilla anguilla*) by sensory chemical and microbiological methods. Food Chemistry, 92: 745-751.
- Losada, V., J. Barros-Velázquez, J. Gallardo and S. Aubourg, 2004. Effect of advanced chilling methods on lipid damage during sardine (*Sardina* pilchardus) storage. Eur. J. Lipid Sci. Technol., 106: 844-850.
- 57. Sikorski, Z.E., J. Olley and S. Kostuch, 1976. Protein changes in frozen fish. Crit. Rev. Food Sci. Nutr.. 8: 97-129.
- 58. Gray, J.I., 1978. Measurement of lipid oxidation: a review. J. Am. Oil. Chem. Soc., 55: 539-546.
- Connell, J.J., P.F. Howgate, I.M. Mackie, H.R. Sanders and G.L. Smith, 1976. Comparison of methods of freshness assessment of wet fish: Part IV. J. Food Technol., 11: 297-308.
- 60. Obemeata, O., F.P. Nnenna and N. Christopher, 2011. Microbiological assessment of stored *Tilapia guineesis*. Afr. J. Food Sci., 5(4): 242-247.
- Bernardez, M., L. Pastoriza, G. Sampedro, J.J.R. Herrera and M.L. Cabo, 2005. Modified method for the analysis of free fatty acids in fish. Journal of Agriculture and Food Chemistry, 53(6): 1903-1906.
- 62. Smith, G., 1988. Lipid oxidation in south East Asian dried fish. Ph.d Thesis, CNAA, Humberside College of higher education.
- 63. Smith, G., S. Hanson and M. Hole, 1988. Lipid oxidation and associated browning in Indonesian salted-dried catfish (*Arius thallasinus*). Fish. Tech. News, pp. 11.
- 64. Burlakova, Y.B., N.M. Storozhuk and N.G. Kharpova, 1988. Relationship between the activity of antioxidants and substrate oxidisabilty in lipids of natural origin. Biophysics, 33: 840-846.
- 65. Virta, S., 2009. Isolation and Identification of Rainbow Trout spoiling Microbiota. Biotechnology and Food Technology, Turku University of Applied Science, pp. 8, Bachelor's Thesis.

- 66. Erkan, N., S.Y. Tosun, S. Ulusoy and G. Uretener, 2011. The use of thyme and laurel essential oil treatments to extend the shelf life of bluefish (*Pomatomus saltatrix*) during storage in Ice. Journal fürVerbraucherschutz und Lebensmittelsicherheit, 6(1): 39-48.
- 67. Mansur, M.A., S. Gheyasuddin and A.K.M.A. Bhuiya, 1989. Preparation of a new ready-to-use dried semi-fermented fish product of increased shelf-life from *Puntiussp.* Bangladesh J. Fish, 19(1): 27-32.
- 68. ICMSF (International Commission on Microbiological Specifications for food) 1986, Recommended microbiological limits for seafoods. In, Microorganisms in Foods. 2. Sampling for Microbiological Analysis: Principles and Specific Applications, 2ndEdn. University of Toronto Press, Buffalo, NY.
- 69. Lilabati, H., W. Vishwanath and M.S. Singh, 1999. Changes in bacterial and fungal quality during storage of smoked, *Esomus danricus* from Manipur. Fishery Technolo, 36(1): 36-39.
- Reilly, A., 1986. Mycotoxins in seafood. In: Cured Fish Production in the Tropics (Reilly, A. and Barile LE, Eds), College of Fisheries, University of Phillipines, pp.131-138.
- Gandotra, R., K. Meenakshi, G. Sweta and S. Shallini, 2012. Change In Proximate Composition and Microbial Count By Low Temperature preservation In Fish Muscle of *Labeo rohita* (Ham-Buch). IOSR J. Pharm. Biol. Sci. (IOSRJPBS), 2(1): 13-17.
- 72. Kolakowska, A., 2002. Lipid oxidation in food systems. In Z. Sikorski and A. Kolakowska (Eds.), Chemical and functional properties of food lipids. London, UK: CRC Press, pp. 133-165.
- 73. Patterson, J. and G. Ranjitha, 2009. Qualities of commercially and experimentally sun dried fin fish *Scomberoides tol.* Afr. J. Food Sci., 3: 299-302.
- 74. Immaculate, J., P. Sinduja and P. Jamila, 2012. Biochemical and microbial qualities of *Sardinella fimbriata* sun dried in different methods. International Food Research Journal, 19(4): 1699-1703.
- 75. Doyle, E.M., 2007. FRI BRIEFINGS: Microbial Food Spoilage: Losses and Control Strategies. *A Brief Review of the Literature*'. Food Research Institute, University of Wisconsin-Madison Http://fri.wisc.edu/docs/pdf/FRIBrief-Microbial-Food Spoilage, pp: 707.