



Biochemical and Microbiological Quality of Three Species of Sundried Fishes of Imphal Market, Manipur

Dr. Hijam Binota Devi

Assistant Professor

Oriental College (Autonomous) Takyel, Imphal, Manipur

Email: bin1971ota@gmail.com

Abstract:

Biochemical and microbiological quality of three sundried fishes namely *Osteobramacotio*, *Puntius sushore* and *Rasbora* sp. available at Imphal Market, Manipur were studied. Average percentages of crude protein, lipid and ash contents were ranged from 48.15 to 57.06, 13.28 to 26.31 and 13.86 to 21.45 respectively. Moisture content was in the range of 10.84 – 14.47 percent. Non protein nitrogen (NPN), soluble protein nitrogen (SPN), total volatile base range of 1.78 – 2.22, 2.97 – 3.20, 39.63 – 64.67, 39.99 – 66.55 and 0.39 – 1.61 respectively. P^H of the fish species varied from 6.49 to 6.63. Total plate count (TPC) for bacteria, total fungal count (TFC), fecal *Streptococci*, *Staphylococcus aureus*, *Bacillus cereus* and most probable number of coliforms were 10³-10⁷g⁻¹, 10-10⁵g⁻¹, 10³-10⁵g⁻¹, 10²-10⁶g⁻¹, 10-10²g⁻¹ and 4.67-10²g⁻¹ respectively. Five different bacterial genera and ten fungal sp. were isolated from the three sundried fishes. Though the present study revealed that the three sundried species were good in quality and within the acceptable limit, proper care should be taken up during processing, handling, transportation and storage to avoid further microbial growth.

Key words: *Biochemical, Microbiological, Sundried Fishes, Imphal Market, Manipur.*

Introduction:

Sundried fishes are widely used by the people of Manipur to enrich flavor of mixed vegetable curry preparations. A large quantity of such cured fishes is also used in the preparations of fermented fishes. As the fish production of the state does not meet the demand, large quantity of sun dried fishes are brought from other neighbouring states and countries. The cured fishes packed in gunny bags reaches Imphal where these are marketed using plywood boxes or split bamboo baskets. The mode of packaging and selling are of great concern in relation to safety and hygienic condition of food

items. Care should be taken up to provide a quality and safety food in relation to the health of the consumers. As there was no reports on quality of such sundried fishes, the present work has to be taken up on the biochemical and microbiological quality of three small sized sundried fish species namely *Osteobramacotio*, *Puntius sophore* and *Rasbora* sp. of Imphal market, Manipur.

Material and Methods:

Sundried fishes namely *O. cotio*, *P. sophore* and *Rasbora* sp. available in Imphal market were collected from different 12 selected fish vendors and brought to the laboratory aseptically in polythene bags for analysis.

Sensory evaluation: Length, weight, reconstitution and organoleptic properties of the fishes were recorded. Lengths of the fishes were measured by using a centimeter scale and weight by a sensitive electric balance. Reconstitution property was assessed as percentage of water imbibed by 100g of the sample soaked in 500 ml of water of a period of 3½ hours as described by Valsan (1975). Organoleptic properties such as colour, texture and odour were recorded; colour was recorded based on simple visual estimation, texture by applying pressure by fingertips on the fishes and odour by smelling the sample. The sundried fishes were grinded and powdered for further analysis.

Biochemical analysis: Total nitrogen (TN), non-protein nitrogen (NPN), soluble protein nitrogen (SPN), moisture, lipid and ash content were determined according to AOAC (1975) methods. Corresponding crude protein value was obtained by multiplying TN values by 6.25. Total volatile base nitrogen (TVBN) and free fatty acid (FFA) was estimated according to the method of Morris (1959). Thio-barbituric acid (TBA) number was determined as per method of Sinhuberand Yu (1958). pH value of the fish samples were estimated according to the method of Valsan (1975).

Microbiological analysis: Enumeration of total viable bacteria (TPC), total fungal count (TFC), most probable number of coliforms and detection of pathogenic bacteria viz. *Salmonella*, *Escherichia coli*, *Staphylococcus aureus*, faecal *Streptococci* and *Bacillus cereus* were done according to the method of APHA (1976). The suspected pathogenic bacteria were further tested as described by APHA (1976) and Kiss (1984). Fungal colonies on PDA were picked, stained with cotton blue in lacto phenol and identified based on Gilman (1957) and Ellis (1971, 1976).

Results and Discussion:

Length, weight, reconstitution and organoleptic properties (colour, texture and odour) of the three sundried fishes were shown in Table 1. Length

ranged from 7.20 to 16.00 cm. for *O. cotio*, 3.50 to 8.80 cm for *P. sophore* and 5.00 to 10.80 cm. for *Rasbora* sp. respectively. Weight varied from 1.80 to 12.47 gm. for *O. cotio*, 0.23 to 4.40 gm. for *P. sophore* and 0.95 to 4.05 gm. for *Rasbora* sp. respectively. Reconstitution properties were in the range of 159.29 to 189.00. The three sundried fishes had crisp to soft texture with varied colours and were of medium to good odour.

Table 2. shows the biochemical composition of the three sundried fishes. Moisture contents of the three fishes were in the range of 10.84 to 14.75 percent. Chakrabarti (1998) also reported the moisture content of small sized dried fishes in the range of 10-13 percent. Sarojnalini and Vishwanath (1994) recorded 18.1 percent moisture content in *P. sophore*, The variation is due to the various physical factors such as temperature, humidity and period of storage of the products. Fish flesh is very susceptible to rapid spoilage in higher ambient moisture content. The main purpose of sun drying of fish is to reduce the moisture content. Crude protein content of the three sundried fishes ranged from 48.15 to 57.06 percent. Highest value was in *Rasbora* sp. Lipid contents of these sundried fish were also high i.e. 13.28 to 26.31 percent. Higher the protein value would be due to the fact that these small sized fishes had no longer exposure to sunlight during drying resulting in the less oxidation process of lipid. Ash content of the fishes were in the range of 13.86 to 21.45 percent. Basuet.al (1989) also reported higher percentage of ash content (11.60 to 24.60 percent) in dry fishes of Andhra Pradesh. Some impurities such as sand and mud during sun drying might also deposited and lead the increased ash composition of the products. NPN and SPN were in the range of 1.78 to 2.22 and 2.97 to 3.20 percent. TVBN, FFA and TBA which were the main indices of spoilage quality ranged from 39.63 to 64.67mg percent, 39.99 to 66.55 percent as oleic acid and 0.39 to 1.61mg malonaldehyde per kilograms. Highest TVBN and TBA were observed in *O. cotio*, FFA in *Rasbora* sp. Sinhuber and Yu (1958) reported that TBA no. less than 3mg per kg. of cured fish product was considered to be in good condition. In the present analysis, the values were within the limit. pH value of the fish samples obtained during the analysis were slightly acidic i.e. 6.49 to 6.63. pH of fish flesh rises from neutrality to 8.0 or even higher as putrefaction proceeds (Reay and Shewan, 1949). In the present analysis, it was below the value.

Bacterial and fungal counts of the sundried fish samples were shown in table 3. TPC for bacteria and TFC for fungi expressed in cfu/g were in the average range of 10^4 to 10^7 and 10^2 to 10^5 respectively. *P. sophore* had the highest count in both cases. Moisture plays important role in the growth of bacteria in fish and lowering of moisture retards the spoilage of fishes (Stanby, 1963). In sundried fish products, moisture level of 20 percent or

below prevents microbial spoilage (Graikoski, 1973) and occurrence of moulds to a magnitude of 10^5 to 10^6 per gram in fish is of great concern. In the present analysis, the total microbial count were not so high and except in *P. sophore* and the values were within the acceptable quality. Even though the present result indicates that these products had some contamination during processing, handling, transportation and storage. But the work of sun drying was well in these fish samples, resulting in lowering the moisture contents below the limit and retarding the microbial growth. However, according to Sillikar (1963), low total count is no assurance of freedom from pathogen. Most probable numbers of coliforms per gram for *O. cotio* and *P. sophore* were 4.67 and 7.2×10 respectively. It was not detected in case of *Rasbora* sp. The occurrence of coliforms in these fishes indicates that they were from an environment having sewage contamination. Faecal *Streptococci* was also detected in these sundried fishes and highest count was obtained in the sundried *P. sophore* (10^5 /g). Presence of these highly resistant enteropathogenic bacteria from these fish samples indicates that these fishes had some faecal contamination. *Staphylococcus aureus* observed in fish samples were in the range of 10^2 to 10^6 per gram. *P. sophore* had the highest count and *Rasbora* sp., the lowest. Iyer (1979) stated that the presence of *Staphylococcus aureus* in fishery products in a few number is not a serious problem but food poisoning may happen if the product is handled carelessly during processing which result in the multiplication of the organism. According to Bergdoll (1979), Staphylococci count exceeding 10^6 per gram is considered to be hazardous. The detected value in the present study resulted that the products were within the limit. Another food poisoning bacteria detected in the fish samples was *Bacillus cereus*. It was detected in *P. sophore* and *Rasbora* sp. in the range of 0.00 to 10^2 and not detected in *O. cotio*. Gillbert et al. (1976 and 1979) reported that *Bacillus cereus* is becoming increasingly important in suspected foods involved in food poisoning. However, incidence of this organism in fishery products within limit is not a serious problem. Other pathogenic bacteria viz. *E. coli* and *Salmonella* were not detected in all the fish samples studied.

The bacterial flora isolated from the three sundried fishes was presented in table 4. five different bacterial genera viz. *Aeromonas*, *Bacillus*, *Flavobacterium*, *Micrococcus* and *Staphylococcus* were detected in the fish samples, out of which four occurred in *O. cotio*, five in *P. sophore* and four in *Rasbora* sp. *Bacillus*, *Flavobacterium* and *Micrococcus* were the common and *Micrococcus* was the dominant genera detected from the fish samples. Some unidentified bacterial genera were also detected. Details of the fungal flora isolated from these sundried fish samples were shown in table 5. About ten fungal species under four genera, one brown sterile and one white sterile

mycelium were identified. The isolated fungal species consisted of *Aspergillus fumigates*, *A. niger*, *A. nidulans*, *Candida* sp., *Cladosporium herbarium*, *Penicillium citrinum*, *P. rugulosum*, *P. sp.*, *P. verruculosum* and *Rhizopus nigricans*. *A. niger* was common to all fish sample. These fungal species detected were airborne species that might come to the products during unhygienic handling, processing, packing, transportation and storage. The sundried fishes are generally poor quality products and unhygienic. The fishes usually have contaminated with other unwanted materials such as sand, dirt, dust etc. during sun-drying. These severely altered in the physical and chemical properties of sundried fishes. However in the present study, the biochemical composition of the three sundried fishes were found to be good. The total microbial count is an indicative of the improved acceptability of the product. In the present result, the total microbial count is low and were within the acceptable limit. However low microbial count could not be considered that the products were free pathogens. These products had contaminations to some extent. So, it could be concluded that to avoid risk of outbreak of diseases, improved drying techniques, proper handling, proper transportation and proper storage should be implemented for good quality products.

Table 1. Reconstitution and Organoleptic Properties of three species of Sundried Fishes

Name of Fishes	Lenth (cm) Texture	Weight (g) Odour	Reconstitution Properties (%)		Colour		
			<i>O. cotio</i>	7.2-16.0 Medium good	1.8-12.47	189±	1.89
<i>P.sophore</i> dry	3.5-8.8 Medium good	0.23-4.4	159.29±	2.22	Dirty white		Soft,
Rasbora sp.	5.0-10.8 Soft	0.95-4.05 Good	165.07±	2.17	Bluish	dorsally,	abdomen pale

Table 2. Biochemical Composition of three species of Sundried Fishes

Composition	<i>O. cotio</i>	<i>P. sophore</i>	<i>Rasbora</i> sp.
Moisture (%)	11.08± 0.29	14.75	± 1.60
10.84 ± 0.09			
Crude protein (%DWB)	55.42 ± 1.89	48.15	± 1.40
27.06 ± 2.44			
Total lipid (%DWB)	11.28 ± 0,39	26.31	± 1.74
24.33 ± 0.48			
Ash (%DWB)	21.45 ± 0.40	19.86	± 0.58
13.86 ± 0.06			
NPN (%DWB)	1.87 ± 0.23	2.22	± 0.49
1.78 ± 0.43			
SPN (%DWB)	3.20 ± 0.23	3.05	± 0.15
2.97 ± 0.62			
TVBN (mg%)	64.67 ± 1.06	39.63	± 1.27
41.00 ± 1.02			
FFA (% as oleic acid)	39.99 ± 0.19	47.35	± 2.23
66.55 ± 2,32			
TBA No. (mg malonaldehyde/kg)	1.61 ± 0.03	0.39	± 0.05
0.97 ± 0.07			
pH	6.53 ± 0.05	6.49	± 0.07
6.63 ± 0.36			

Note: Results are Mean ± S.D. of 12 samplings

DWB=Dry Weight Basis, NPN=Non Protein Nitrogen, SPN=soluble protein nitrogen,

FFA= Free fatty Acid, TVBN= Total Volatile Base Nitrogen, TBA= Thio-barbituric acid.

Table 3. Bacterial and Fungal Count of three species Sundried fishes

Name of fishes		TPC/g Coliforms (MPN/g) (Bacterial) (MPN/g)	TFC/g (Fungi)	Faecal <i>Streptoco- ccus/g</i>	<i>Staphylo- coccus aureus/g</i>	<i>Bacillus cereus/g</i>
<i>O. cotio</i>	Mean	4.97x10 ⁴	1.07x10 ²	6.33x10 ³	2.10x10 ³	ND
	4.67					
	Range	1.00x10 ⁴ -	3.00x10 ¹ -	1.00x10 ³ -	8.00x10 ² -	
	9.10-	7.8x10 ⁴	2x10 ²	1.20x10 ⁴	3x10 ³	
		2.30x10 ¹				
<i>P. sophore</i>	Mean	2.06x10 ⁷	1.26x10 ⁵	8.63x10 ⁵	1.48x10 ⁶	1.05x10 ²
	7.2x10 ¹					
	Range	2x10 ⁵ -	2.00x10 ² -	1.00x10 ⁵ -	3.90x10 ² -	1.00x10 ¹ -
	1.2x10 ² -	9.8x10 ⁷	3.00x10 ⁵	4.15x10 ⁶	3.30x10 ⁶	2.00x10 ²
		3.60x10 ²				
<i>Rasbora sp.</i>	Mean	1.25x10 ⁴	1.89x10 ²	5.42x10 ³	1.55x10 ³	1.00x10 ¹
	ND					
	Range	1.02x10 ³ -	1.00x10 ¹ -	1.20x10 ³ -	8x10 ² -	0.00-
		1.47x10 ⁴	2.74x10 ²	1x10 ⁴	1x10 ⁴	2x10 ³

Note: Results are Mean ± S.D. of 12 samplings. ND = not detected

Table 4. Bacterial flora of three species of Sundried fishes (%).

Bacteria	<i>O. cotio</i>	<i>P. sophore</i>	<i>Rasbora sp.</i>
<i>Aeromonas</i>	0.00-1.45	8.16-10.24	ND
<i>Bacillus</i>	1.00-8.69	1.00-5.56	0.00-2.23
<i>Flavobacterium</i>	0.00-14.28	4.17-8.33	1.32-3.33
<i>Micrococcus</i>	1.45-37.12	1.02-31.48	2.13-25.40
<i>Staphylococcus</i>	ND	0.00-4.73	4.27-6.03
Unidentified	11.00-33.33	17.00-30.39	10.38-25.00

ND = not detected

Table 5. Fungal flora isolated from three species of Sundried fishes

Microflora	<i>O. cotio</i>	<i>P. sophore</i>	<i>Rasbora sp.</i>
<i>Aspergillus fumigates</i>	+	-	+
<i>A. Niger</i>	+	+	+
<i>A. sidowi</i>	-	+	+
Brown sterile mycelium	-	+	-
<i>Candida sp.</i>	-	+	+
<i>Cladosporiumherbarum</i>	+	-	-
<i>Penicilliumcitrinum</i>	+	-	+
<i>P. rugulosum</i>	-	+	-
<i>P. sp.</i>	-	+	+
<i>P. verruculosum</i>	-	+	-
<i>Rhizopus nigricans</i>	-	+	+
White sterile mycelium	+	+	+

+ present; - absent

References:

1. AOAC (1975). *Official Methods of Analysis* (12th edn.). Association of official Analytical Chemists, Washington D.C.
2. APHA (1976). *Compendium of Methods for Microbiological Examination of Foods*, ed., M.L. Speak, American Public Health Assn., Washington, 701.
3. Basu, S. D., Imam Kashim, S. S. Gupta and C. C. Panduranga Rao. (1989). Quality of Dry Fish from Markets in Andhra Pradesh. **26**, 114-118.
4. Bergdoll, M. S. (1979). *Foodborn Infections and Intoxications* (eds. Rieman, H. and Bryan, E). Academic Press, New York, U.S.A., 444
5. Chakrabati, R. (1998). Non Traditional Products from small Fish. *J. Fishing Chime*, **18**(7), 52-54.
6. Ellis, M.B. (1971). *Dermateceous Hypomyces*, **CMI**, Kew, Surrey, England, 608.
7. Ellis, M.B. (1976). *More Dermateceous Hypomyces*, **CMI**, Kew, Surrey, England, 507.
8. Gilbert, R. J. (1979). In *Food borne Infections and Intoxications*, Rieman, H. and Bryne, F. L. (eds.), Academic Press Inc., 495.
9. Gilbert, R. J. and Taylor, A. J., (1976). In *Microbiology in Agriculture, Fisheries and Food*, Skinner, F. A. and Carr, J. G. eds. Symposium series No. 4 of the society for Applied Bacterial, Academic Press, 197.
10. Gilman, J. C. (1957). *A manual of Soil Fungi*, The Iowa State University Press, Iowa, U.S.A. Grraikoski, J. T. (1973). Microbiology of cured and fermented fish. In *Microbial. Safety of Fishery Products*, eds. C. O. Chichester and H. D. Graham. Academic Press, New York, 97-112.
11. Graikoski, J.T. (1973). Microbiology of cured and fermented fish. In *Microbial. Safety of Fishery Products*, eds. C.O. Chichester and H.D. Graham. Academic Press, New York, 97-112.
12. Iyer, T.S.G. (1979). Pathogens and faecal indicator organisms in fish and fishery products. In *quality Control in Fish Processing*. Central Institute of Fish Technol., Cochin (ICAR), 45-59.
13. Kiss, I. (1984). *Testing methods in food microbiology*. Elsevier Asterdam Oxford New York, Tokyo, 437.
14. Lilabati, H. and Vishwanath, W. (2000). Biochemical, nutritional and microbiological quality of six species of smoked fishes of Manipur. *J. Fish. Technol.* **37** (2), 89-94.
15. Morris, B. J. (1959). *The Chemical Analysis of Foods and Food Products*. D. Van Nostrand Co. Inc. Princeton, New Jersey, 970.

16. Reay, G.A. and Shewan, J.M. (1949). The spoilage of fish and its preservation by chilling. *Adv. Fd. Res.* **11**, 343 – 398.
17. Sarojnalini, C. and Vishwanath, W. (1994). Composition and Nutritive value of sundried *Puntius sophore*. *J. Fd. Sc. Technol.* **31**(6), 480-483.
18. Sillikar, J.H. (1963). Total Count as Index of Food Quality. In *Microbiol Quality of Foods*. Eds. L.W. Slanetz, C.O. Chichester, A. R. Ganfin and Z.I. Ordal, Academic Press, New York, London, 273.
19. Singh, M.B. Sarojnalini, C. and Vishwanath, W. (1990). Nutritive value of sundried *Esomus danricus* and smoked *Lepidocephalus guntea*. *J. Food Chemistry.* **36**, 89-96.
20. Sinhuber, R.O. and Yu, T.C. (1958). 2 – Thiobarbituric acid method for the measurement of rancidity in fishery products. II. Qualitative Determination of Malonaldehyde. *Fd. Technol.*, **12** (1), 9-12.
21. Stanby, M.E. (1963). Cured fishery products. In *Industrial Fishery Technology*, eds. M.E. Stanby and E. Robert, Krieger Publ. Co. Hunligton, New York, 415.
22. Valsan, A.P. (1976). A comparative yield and biochemical evaluation of the existing fish curing methods in India. *Proc. Symposium of Fish Processing Industry in India*. AFST and CEFTRI, Mysore, 77-79.