Chapter VI

Summary & Conclusion
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The people of Manipur depend mainly on fish for their animal protein requirement. Fish production of the state, however, does not meet the demand of the growing population. Thus, large quantities of iced and smoked fishes are purchased from other states and sold in Manipur. Muscle of fresh fish is rich nutritionally and is considered sterile. But, contaminations occur during transport, preservation and processing. Nutritional quality of the fishes may consequently change. The quality of the fishes need be checked regularly with a view to maintain human health and hygiene. As there is no much work done so far on the microbiological and nutritional quality of the fishes sold in Manipur, the present work was undertaken with the following objectives:

2. Studies on the nutritional quality of the fishes.
3. Studies on the microbiological quality of the fishes with special emphasis on the identification of pathogenic organisms.
4. Studies on the microflora associated with the fishes.

Fish samples examined were:

**Fresh fish:** six fresh fishes, viz, *Clarias batrachus, Wallago attu, Labeo rohita, Anabas testudineus, Lepidocephalus guntea* and *Monopterus albus* were
taken. These fishes were from the water bodies of Manipur valley and sold in the Imphal market of Manipur.

**Iced Fish:** Six iced fishes, viz., *Wallago attu*, *Labeo rohita*, *L. gonius*, *Aorichthys aor*, *Notopterus chitala* and *Hilsa ilisha* were taken. The fishes were transported from other states and sold in the Imphal market.

**Smoked Fish:** Six smoked fishes, viz., *Clarias batrachus*, *Wallago attu*, *Anabas testudineus*, *Channa punctatus*, *Tor putitora* and *Puntius jayarami* were taken. *Clarias* was collected from the Imphal market (caught and smoked in Manipur), the next three from Assam and the last two were from Moreh town of Manipur bordering with Myanmar.

**BIOCHEMICAL COMPOSITION, SPOILAGE AND SENSORY EVALUATION:**

**Fresh Fish:** Crude protein, lipid, ash and moisture contents of the fishes were (per cent dry weight basis DWB), 70.00-81.8, 5.74-12.26, 4.00-13.00 and 75.00-79.43 respectively. Highest value of protein was in *Wallago attu*, lipid in *Anabas testudineus* and Ash in *L. guntea*. NPN (Non protein nitrogen) value was 1.61-2.54 %, highest being in *L. rohita* and lowest in *Anabas testudineus*. *Wallago attu* has highest TVBN (Total volatile base N) value, 11.00 mg %. pH was 6.58-6.90. Sensory tests of all the fresh fishes were in excellent condition.

**Iced fish:** Crude protein, lipid, ash and moisture contents of the fishes were (% DWB), 70.38-84.59, 5.91-20.62, 3.30-4.01 and 78.00-80.47 respectively. Highest value of protein, lipid and ash were in *L. gonius*, *A. aor* and *N. chitala*.
respectively. NPN value was highest in *H. ilisha*. In all the fishes examined, fairly high TVBN values of 12-14 mg% were observed. FFA (free fatty acid) values and TBA (thiobarbituric acid) number were high in *N. chitala* and *W. attu*. It indicates that lipid hydrolysis and oxidation were more in the fish. Muscle pH values were 6.4-6.9 in all the fishes.

Sensory test of *A. aor* was adjudged good and other fishes were adjudged fair. Some specimens of *W. attu* and *H. ilisha* had slightly rancid odour.

**Smoked fish:** Moisture contents of the smoked fishes brought from outside the state were low (6.0-15.0%) compared to that of *Clarias* (47.6-6.0%) of Manipur. Protein, lipid and ash contents were (%DWB) 71.44-83.40, 3.70-10.73 and 4.00-9.41 respectively. Highest value of protein was in *P. jayarami*, lipid in *T. putitora* and ash in *C. punctatus*. NPN varied from 0.83 to 4.85%, highest was in *T. putitora*. *W. attu* had the highest TVBN (39.00mg%) and FFA (8.65% oleic acid) values. Smoked fishes were slightly acidic within the range of pH, 6.30-6.40.

Reconstitution properties were highest in *C. batrachus* (194.00%) and lowest in *T. putitora* (90.00%). Most of the fishes had hard texture except *C. batrachus* smoked in Manipuri style. *C. batrachus, P. jayarami* and *T. putitora* had pleasant smoky odour and *A. testudineus, W. attu* and *C. punctatus* had medium smoky odour.

**NUTRITIONAL QUALITY OF FISHES**

**Fresh fish:** *In vivo* feeding experiments were conducted using male albino rats. Rats fed on *M. albus* had highest growth rates (129.33 gm). PER (Protein efficiency ratio) values of the fishes were 2.22-2.60. *M. albus* had the highest value,
while reference casein had 2.44. In all the fishes protein digestibility values were above 83%. In all the fishes, protein digestibility values were above 83% and (BV) Biological values were above 90.00%. Highest digestible and BV were observed in C. batrachus and A. testudineus.

**Iced fish:** Highest growth rate was observed on the rats (102.01 gm) fed with H. ilisha. PER value of the fishes were 2.12-2.62, highest beings in H. ilisha. Digestibility values were above 80%, highest was in H. ilisha. B. V. were also above 90%, the highest in H. ilisha.

**Smoked fish:** Digestibility values in the pepsin phase were, 73.87-89.63% and in the trypsin phase were 82.70-91.50%. Both in pepsin and trypsin phases, highest digestibility was of W. attu. Alpha amino N liberated during enzyme hydrolysis both in pepsin and trypsin phase, was highest in C. batrachus. Alpha amino constituted 50.88% of total nitrogen in pepsin phase and 62.46% of total nitrogen in trypsin phase.

**MICROBIOLOGICAL QUALITY AND TEST FOR PATHOGENIC BACTERIA.**

**Fresh fish:** Bacterial counts ranged from $10^4$-10$^7$ cfu/g. Highest was in M. albus (5.5x10$^6$) and lowest in L. guntea (9.0x10$^4$). C. batrachus had the highest fungal count (2.8x10$^3$ cfu/g) and L. guntea had the lowest count (7.0 cfu/g). Coliform, Staphylococcus aureus, faecal Streptococci were observed in all the fishes, but E. coli and Salmonella were not detected.
**Iced fish:** In most of the fishes, highest count was observed in the gill and lowest in the muscle. The count in skin was variable. TPC/g or cm² ranged from $10^5$-$10^9$ cfu, highest being in *A. odor* ($10^9$ cfu). TFC/g or cm² was ranged from $10^2$-$10^4$ cfu; highest was observed in *N. chitala* ($10^4$ cfu). Coliform, *Staphylococcus aureus*, faecal *Streptococci* were observed in all the tissue of the sample analysed, but *E. coli* and *Salmonella* were not detected.

**Smoked fish:** TPC/g and TFC/g in smoked fishes ranged from $10^5$-$10^{10}$ cfu and $10^2$-$10^5$ cfu. Highest count of TPC was in *P. jayarami* ($1.0 \times 10^{10}$ cfu) and TFC in *C. batrachus* ($2.9 \times 10^5$). *Staphylococcus aureus*, faecal *Streptococci* and *Bacillus* were detected in all the samples, but here also *E. coli*, *Salmonella* and coliforms were not detected.

**MICROFLORA ASSOCIATED WITH THE FISH**

**Fresh fish:** Most of the bacteria isolated were gram +ve cocci and gram -ve rods. The number of genera isolated from *C. batrachus, W. attu, L. rohita, L. guntea, M. albus* and *A. testudineus* was 4, 5, 5, 4, 5 and 7 respectively. The bacterial flora were *Acinetobacter, Aeromonas, Micrococcus, Moraxella, Staphylococcus, Pseudomonas* and *Enterobacteriaceae*. Some colonies could not be identified. The common bacterial flora were *Aeromonas, Micrococcus* and *Staphylococcus*. The number of the genera of fungi isolated from *C. batrachus, W. attu, L. rohita, L. guntea, M. albus* and *A. testudineus* were 4, 4, 5, 3, 4 and 3 respectively. Isolated flora were *Candida, Fusarium, Cladosporium, Rhizopus,*
Aspergillus, Penicillium and Gleosporium. Some were sterile mycelia and some could not identified. The common fungal genera were Aspergillus and Penicillium.

_Iced fish:_ The bacteria isolated from iced fishes were Acinetobacter, Aeromonas, Flavobacterium, Micrococcus, Moraxella, Pseudomonas, Vibrio, Photobacterium, Bacillus, Staphylococcus, Lactobacillus and Enterobacteriaceae. Micrococcus, Aeromonas, Pseudomonas and Flavobacterium were found in higher percentage. Ten genera of fungi were identified and some was sterile mycelia on potato dextrose agar (PDA). They were Aspergillus, Cladosporium, Fusarium, Geotrichum, Mucor, Penicillium, Rhizopus, Phoma, Geocladium and Trichoderma. Count of Fusarium, Penicillium and Aspergillus were found in higher percentage.

_Smoked fish:_ Bacteria isolated from smoked fishes were Bacillus, Staphylococcus, Micrococcus, Xanthomonas, Pseudomonas and Streptococcus. Some colonies could not be identified. In most of the fishes, Bacillus and Micrococcus were found in higher percentage. Five genera of fungi were identified. They were Aspergillus, Penicillium, Rhizopus, Candida, and Cladosporium. Aspergillus and Penicillium were the two genera which were predominantly found in smoked fishes.

From the above result it may lead to the conclusion that inspite of high bacterial and fungal counts. Fresh, iced and smoked fishes of Manipur may be recommended for human consumption as the fishes examined were in acceptable condition, highly nutritive and were devoid of common harmful bacteria.
However, the quality of the fishes could be improved by taking up certain measures as follows:

1. After harvesting, fish should not be exposed to direct sunlight or to wind. However, it should be carefully cleaned and cooled down to 0°C as quickly as possible. Washing reduce the microbial load by 80-90%. Delay in icing reduces the shelf life of the fish.

2. Processing should be done carefully especially to maintain sanitary conditions. Water supply for use in processing and ice manufacture must be chlorinated. Processing rooms must be regularly cleaned to keep down bacterial contamination. Persons engaged in processing should be neat and clean.

3. Repeated icing, thawing and spontaneous exposure to atmosphere should be prevented. It causes gradual increase in microbial loads and also leads to considerable losses of nutrient rich tissue water as drip and also lowers the juiciness and hence general acceptability.

4. Establishment of re-icing facilities on the way to Manipur, establishment of proper cold storage and processing facilities in the state may improve the quality of iced fishes.

5. Strict care during selection of fresh fish, processing before icing will contribute to the maintenance of quality at the marketing site.

6. Processing requires knowledge of the microbiology of the raw product. This is of vital importance in food processing, since micro-organisms surviving the
processing condition will reflect the microflora of the original product as well as that obtained in handling and storage.

7. The smoked fishes may be consumed after proper sterilization, either by roasting or cooking. As micro-organisms readily propagate in fishes even after smoking, proper sanitary care should be taken during and after processing of the fishes.

8. The improved technique of smoking should be adopted. If it is processed in a quality controlled and well-designed kiln, it could prevent the loss of nutritive value and also help villagers to produce better processed products in large scales. The selection of good raw materials is also very important for getting a good product.

To improve the quality of this particular popular item, the state Fisheries Department and other volunteer organizations engaged in this business should come forward to take up corrective measures regarding the way of selling, mode of transportation etc. in order to serve the general fish eating mass of Manipur.