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## Basics of sample collection, preservation and species identification of finfish

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### Introduction

Fisheries are one of the most important renewable resources. With increasing fishing pressure, the only option left for the sustainability of fisheries is their rational management. Proper management is possible with a thorough knowledge of the dynamics of the fish stocks. For a meaningful study of the dynamics, knowledge of natural history of the species is necessary and this in turn can be acquired by the correct identification of fish species. This assumes greater importance in tropical seas where, a multitude of closely related and morphologically similar species occur. The role of taxonomy and proper identification cannot be overstressed in studies of population dynamics. Acquaintance with the main species should be such that there should no errors in identification of them in any special form such as racial differentiation, abnormalities, malformation due to decay or disease. As to species of less importance collections and observations can be made for taxonomic studies which will be useful in future. Species identification study is also a step towards understanding the bewildering biodiversity that characterizes in the marine ecosystem.

Pioneering studies on taxonomy of Indian fishes began in the late 18th century by European scientists and officers of the British East India Company. One of the pioneers was Bloch (1795), and his student Schneider (1801), followed by Lacepède (1798-1803). Dr. Alcock who undertook the first marine fisheries survey in India published the findings in 1869. Perhaps the most important work during this period, pertaining to the subject was that of Sir Francis Day, Surgeon Major with the British troops in Bengal, who studied the systematic of Indian fishes in depth for over 20 years. His monumental work was published in two volumes as the 'The Fishes of India: being a natural history of the fishes known to inhabit the seas and freshwaters of India. Vol.I and II' (1878) and the 'The Fauna of British India, including Ceylon and Burma' (1889). During the subsequent period of one century, a

large number of fishes have been described and added to the list already prepared by Day, and the important works during this period with regard to the taxonomy of fishes of the Indian waters are by Munro (1955), Jones and Kumaran (1980), who published descriptions of over 600 species from Laccadives archipelago, Talwar and Kacker (1984), and the most recent compilation is that of Talwar and Jhingran (1991a, 1991b), who published descriptions of a total of 930 species of inland (fresh and brackish water) fishes of India.

### i. Sampling and Preservation

#### Sampling for taxonomic studies

For taxonomic study, specimens of all species occurring in the ecosystem covering the entire length range were collected and preserved in 5% formalin after injecting 5% formalin through the vent and dorsal musculature. The specimens were preserved in a wide mouthed bottle in such a manner that the shape is not distorted during storage. Morphometric and meristic data were taken following Hubbs and Lagler (1947). Measuring linear dimensions of whole or parts of fish is probably the most widely used technique in taxonomic studies. Such observations are made with taps and calipers. Measurements are usually but not always taken along straight lines.

#### Fish Collection Methods

The major objective of the bioinventory is to identify all the available species in the habitat using all the gear combinations. Two types of gears are employed can be divided in to two viz, active and passive categories. Passive gear is usually set and left stationary for a period and commonly used gear are gillnet and traps. Active gears used in the inventory are seine nets, trawl nets, dip nets, hooks and line and electric fishing. Different factors affect fish sampling such as water depth, conductivity, water clarity, water temperature, fish size and fish behavior.

## Fish Preservation techniques

Careful and correct preservation procedures are important for getting good results of collected specimens. Usage of correct fixative, correct concentration, appropriate containers, clean and sharp dissecting tools, quality labels will enhance the quality and value of the preserved specimens. Voucher specimens are the representative samples of species identified in the field and preserved to verify the field identification. Type specimens are the name bearing specimen preserved to verify the taxonomy of species.

### Preservation

All fish must be killed before the fixation and preservation. This can be achieved by keeping the fish in high doses of the anaesthetizing solution like clove oil; killing of fish is an ethical treatment of a live animal and also serves scientific purpose. Make a small incision on the right side of the abdomen to facilitate penetration of preservative. The right side is chosen for this and similar operations like removal of scale samples, because left side is used for getting morphometric data and is commonly shown in photographs.

Time of the formalin fixation of the average specimen may range from two days to a week. For permanent preservation formaldehyde can be removed by soaking the specimen at least for two days in fresh water with regular water change. The permanent preservation can be done in 70% of alcohol or 40 % alcohol.

### Fixation

- (1) Formalin: It is commonly used preservative for specimens and it is available in the liquid form. About 10% of formalin is used for preservation of fish specimens. 10% solution of buffered formalin is prepared by combining in 1 part full strength formalin and 9 parts distilled water and add 3ml of borax per litre.
- (2) Paraformaldehyde: It is in a powder form and can be used to make formalin solution. A mixture of 1 part paraformaldehyde, 4 parts of anhydrous sodium carbonate and a small amount of Alconox (wetting agent) can be added to 20g powder mixture of 400ml of distilled water to produce 10% buffered formalin solution.
- (3) Alcohol: Ethanol and Isopropanol are commonly used to fix and preserve fish specimens.

### Fixation procedure

Specimen should be fixed immediately after collection. To fix the specimen wide mouthed glass filled with fixative solution is can be used. Tight lids must be used for fixation. The specimen must be preserved in as natural as possible. The fixative should be injected to the body cavity to facilitate penetration and preservation of internal organs.

## Genetic sampling

Recently more attention is being done to utilize genetic markers for population studies. These techniques are useful in fisheries management to identify populations of fish, select broodstock, assess purity of hatchery stock, determine genetic population structure and assess biodiversity at the genetic level. Commonly used techniques are electrophoresis and DNA analysis.

Tissue samples can be collected from muscle tissue, fin clips, scales and epithelial tissue.

Samples classified in to three categories depending upon the method utilized-

- (1) Protein electrophoresis: Requires organ tissue including heart, muscle, eye, and kidney, either fresh or fresh frozen.
- (2) DNA analyses with no PCR amplification: Requires large amounts of tissue often fresh and in buffer solution.
- (3) DNA analyses with PCR amplification: Requires very little tissue (fin clips, scale) and can be preserved in ethanol.

### Labelling

Labelling has to be done neatly and should not make any confusion in the future. Labels include date of collection, place of collection, ecological details, method of capture and name of collector.

## ii. Identification of fish

To learn the characters of importance for the identification of fishes and by which they may be accurately identified. Line drawings, colour plates and photographs provide basis for the learning the salient characters used for their classification. Identification keys can be used as distinguishing characters of each family and order in which forms are treated according to the phylogeny.

### Example:

1. Determine the family in "Key" to the families".
2. Identify to the lowest taxonomic unit listed in key to the family of which the fish is member
3. Verify the final determination by ascertaining -by comparing the similarities of the specimen with illustration.
4. The specimen collected matches specimens previously identified by taxonomist.
5. Confirm the geographical range as given in the standard texts includes the locality from which the specimen was taken.
6. Compare the descriptions given in the FAO identification sheets, Smiths sea fishes, Catalog of Fishes and Fish base.

### iii. Measurements

Smoothly working dividers or digital calipers can be used for measurements. A steel scale of good quality is recommended for precise reading. Measuring board commonly used in fishery biology investigations is not suitable for taxonomic studies. All measurements are taken in a straight line

**Definition of Body Measurements** (All measurements along the antero-posterior axis)

1. **Total Length (TOL):** The greatest dimension between the most anteriorly projecting part of the head and the farthest tip of the caudal fin when the caudal rays are spread out together.
2. **Standard Length (STL):** The distance from the anterior most part of the head backward to the end of the vertebral column (structural base of caudal rays).
3. **Fork Length (FOL):** Distance from the tip of snout to the end of the middle ray of the caudal fork when the fish is being flattened out.
4. **Head Length (HEL):** Taken from the tip of the snout to the posterior most point reached by the bony margin of the operculum.
5. **Pre-orbital length (PRO):** Distance from the tip of the snout to the forward point of eye.
6. **Eye diameter (EYD):** Horizontal diameter of the visible part of the eye, i.e., the distance between the front edge and the back edge of the orbit.
7. **Postorbital length (PSO):** Distance from the backward point of eye to middle of the backward bony edge of the operculum.
8. **Upper jaw length (UPJ):** Length of maxillary is taken from the anterior most point of the premaxillary to the posterior point of the maxilla.
9. **Lower jaw length (LOJ):** Length of lower jaw from anterior tip to angle of mouth.
10. **Body depth (BDD):** Distance between the middle point of dorsal finbase to straight downward central margin of the body, excluding fins.
11. **Pre-dorsal length 1 (PD1):** Distance from the tip of the snout to the forward origin of the dorsal (intersection point of the forward edge of the first ray of the dorsal, D1, with the outline of the back, the fish being flattened out)
12. **Pre-dorsal length (PD2):** Distance from the tip of snout to the forward origin of the dorsal (intersection point of the forward edge of the first ray of the dorsal, D2, with the outline of the back, the fish being flattened out).
13. **Pectoral fin length (PEL):** Distance from the extreme base of the uppermost ray to the farthest tip of the fin, filament if any.
14. **Pelvic fin length (PVL):** Distance from the extreme base of the uppermost ray to the farthest tip of the fin, filament if any.
15. **Dorsal fin length 1 (DF1):** Distance from the origin of the tip of the fin to the anterior lobe.
16. **Dorsal fin length 2 (DF2):** Distance from the origin of the tip of the fin to the anterior lobe.
17. **Inter dorsal length (IDL):** Distance from the base of the last spine (ray) of first dorsal to the intersection point of second dorsal fin.
18. **Pectoral fin base length (PEB):** Distance from the base of the anterior fin ray of the pectoral (P) to the backward end of the last ray, the pectoral being extended on the side of the fish in its normal position.
19. **Pelvic fin base length (PVB):** Distance from the base of the anterior fin ray of the pelvic fin (P) to the backward end of the last ray, the ray being extended on the side of the fish in its normal position.
20. **Dorsal fin base length (DB1):** Distance from the forward origin of the dorsal (D1) to the backward edge (Intersection point of the backward edge of the last spine, D', with the outline of the back, the fin being extended).
21. **Dorsal fin base length (DB2):** Distance from the forward origin of the dorsal (D2) to the backward edge (Intersection point of the backward edge of the last ray, D2, with the outline of the back, the fin being extended).
22. **Anal fin length (AFL):** Distance from the origin of the tip of the fin to the anterior most outer tip of the anal fin.
23. **Anal fin base length (ABL):** Distance from the forward origin of the anal (A) to its backward edge (intersection point of the backward edge of the last ray, A' with outline of the abdomen, the fin being extended).
24. **Caudal peduncle length (CPL):** Distance from the base of the second dorsal end to origin of the caudal fin.
25. **Caudal peduncle depth (CPD):** Depth of the caudal peduncle.
26. **Pre-pelvic length (PRP):** Distance from the tip of the snout to the anterior origin of the pelvic (intersection point of the forward edge of the first ray of the pelvic, with the contour of the abdomen, the fin being extended).
27. **Pre-pectoral length (PRV):** Distance from the tip of the snout to the margin of the insertion of pectoral fin.
28. **Pre-anal distance (PRA):** Distance from the tip of the snout to the forward origin of the anal (interior point of the forward edge of the first ray of the anal, A, with the outline of the abdomen, the fin being extended).

### iv. Description

**Description of specimens:** Detailed biological analysis of specimens have to be made and data to be recorded.

**Morphology:** General morphology, appearance, body shape, depth contour and nature of fins.

**Colour:** Fresh as well as preserved colour pattern, spots, markings, colour changes in the larvae and adult.

**Scales:** Nature of scales and description.

**Reproductive system:** Structure of reproductive organs of

female and male. Description of different parts, nature of gonads, stages of maturity, nature of egg.

**Digestive system:** Nature, length of alimentary canal, structure of alimentary canal, if any modifications of stomach and intestine.

### Suggested Reading

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