GENERAL DIRECTIONS FOR LABORATORY STUDY

The main objective of the laboratory work is to verify what a student has learnt in the laboratory and to develop a sense of keen observation and confidence. Before coming to the laboratory the students are advised to be fully prepared with the theoretical knowledge of the experiment/ exercise of the day. The students are required to come with all the essential equipment pertains to the lab work. The students should listen carefully the instructions given by their teacher and try to develop a habit of removing their difficulties or doubts from their teachers.

(a). Museum Study:

1. First of all observe and study the museum specimen to be drawn.
2. Draw directly from the museum specimen and complete your all drawings in the laboratory. Label them with the help of some text book or practical manual.
3. Draw dorsal, ventral or lateral views of the specimen.
4. Ratio proportion of the original specimen should always be kept in mind.
5. Draw always line diagrams, avoid shading and never give an artistic touch to them.
6. Never use colored pencils and ink.

(b). Study of Slides:

1. In the case of permanent slide, give a well labeled and neat diagram showing details of entire slide or a part of it.
2. Mention the important points for its identification.
3. If the slide has a mount of entire animal, mention its classification also.

(c). Dissection:

1. Before dissecting the animal listen the instructions given by the teacher and follow them strictly throughout the dissection. Keep patience during the dissection and never dissect the animal until your are sure what you are going to do.
2. The animal or its part to be dissected should be kept moist at all times. The specimen should be placed or pinned in the dissecting tray. The pins should be fixed obliquely.
3. All vertebrates should be dissected ventrally and all invertebrates from the dorsal side.
4. The waste should not be thrown carelessly on the table or floor, keep it either in the dissecting tray or near by sink slowly.
5. Change the water time to time so as to remove the turbidity caused by body muscles and blood etc.,
6. During the dissection muscles should be separated parallel to the fibers where as the blood vessels and nerves along and not across. Always keep the scissors parallel to the longitudinal axis.

7. A well labeled diagram of the dissection should be drawn on the practical note book.

8. The dissection should be nicely and correctly displayed with the help of black and white papers and flag labels, if necessary.

(d). Practical Note Book or Record:

An upto date practical note book with correctly drawn and labeled diagrams and duly signed by the teacher should be presented. A table of contents should be given in the beginning of the note book.

MUSEUM STUDY

The study of museum includes the preserved animals, stuffed animals and models in the museum. The students are advised to observe the specimens carefully in the laboratory. The students should note the characteristic features of the specimens provided in the laboratory. After careful examination students should draw a neat and clean diagram of the specimen. These diagrams must be labeled. The labeling must be reasonable. Write the name of the museum specimen at the bottom and systematic position on the right side of the specimen whether solitary, colonial or whole mount on the slide. The comments are the most important things in spotting. The students should mention reasons for the classification. i.e. at least one important and distinguishing character of phylum, group, sub-phylum, class, sub-class, order and sub-order etc.,. After it mention the habits and habitat of the animal, special morphological features.

### 1. Identification of fish and prawn (museum study)

i).Scoliodon
Classification

Phylum	-	Chordata
Group	-	Craniata
Subphylum	-	Vertebrata
Division	-	Gnathostomata
Superclass	-	Pisces
Class	-	Chondrichthyes (Elasmobranchi)

subclass	-	Selachi
Order	-	Pleurotremata
		(=Squali)
family	-	Scyllidae
Genus	-	*Scoliodon* (Dogfish)

Habit and habitat. The natural home of Scoliodon is the sea, but some live in **estuaries** and even ascend the rivers. They are predacious and voracious feeders attacking their prey with powerful jaws. They are active swimmers.
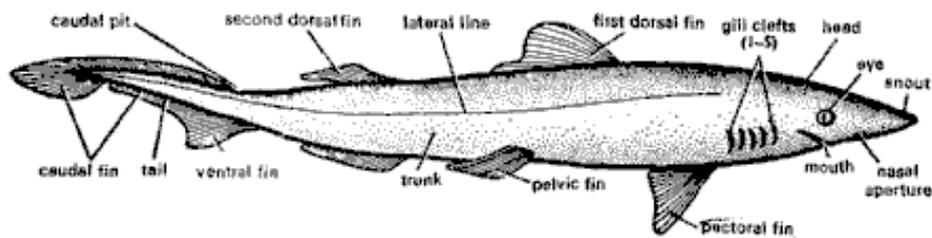
Comments:

(1) Scoliodon is commonly called **dogfish** or **dog shark**.
(2) Spindle-shaped body, about 60 cm long, is regionated into head, trunk and tail.
(3) Dorsal and lateral sides of body are pigmented dark grey or slaty grey, while the ventral side is white.
(4) Head is dorso-ventrally compressed and flattened into snout. It contains ventrally situated slit-like mouth, obliquely situated nostrils and laterally situated protuberant

eyes. A little behind eyes there are five pairs of lateral gill-clefts.

(5) Trunk bears (I) median unpaired and (ii) lateral paired fins. Median unpaired fins are (a) large first dorsal fin, (b) small second dorsal fin, and (c) ventral fin. Paried fins include a pair of anterior pectoral fins and a pair of posterior pelvic fins.

(6) Heterocercal tail containing musculature and vertebral column is turned upwards.

(7) A pair of pigmented lateral lines extends from head to tail.

(8) Scoliodon exhibits sexual dimorphism. Males are easily recognized by having a pair of intromittent organs, called as claspers. Cloaca is found between 2 plevic fins.

**Identification.** Since the animal has raised tail, pointed snout, and above features, hence it is *Scoliodon*.

ii). Sphyrna (Hammer-headed Shark)

Classification. Same as in Scoliodon.

Habit and habit. Sphyrna or Zygaena or Reniceps is a common marine fish, adapted for deep sea waters. It is a voracious feeder and active swimmer. It eats small fishes, but because of its attacks on man, it is dreaded as man-eater.

Comments:

(1) Elongated body measuring 4-5 metres and divided into head, trunk and tail.
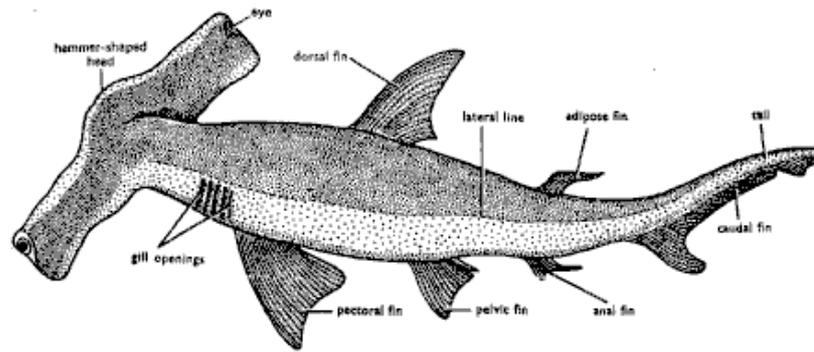
(2) It is called hammer-headed shark due to the hammer-shaped head, which is produced into two prominent lateral lobes supported by corresponding cartrilaginous outgrowth from post-orbital or lateral ethmoidal region of skull. Eyes containing nictitating membrane are placed at the distal extremities of the lateral lobes. Lateral expansions of head, with eyes and nasal openings, farther apart than sharks, confer an advantages in homing in on food.

(3) First dorsal fin is situated in front of pelvic fin and second dorsal fin opposite to anal fin and both devoid of spines.

(4) Mouth is crescentic and ventral in position.

(5) Gill-slits 5 pairs and lateral in position. Spiracles are absent.

Identification. Since this fish has hammer-shaped head and above features, hence it is Sphyrna.

iii). Torpedo (Electric Ray)
Classification

Phylum   - Chordata
Group   - Craniata
Subphylum   - Vertebrata
Division   - Gnathostomata
Superclass   - Piscese
Class   - Chondrichthyes
Subclass   - Selachi
order   - Hypotremata
family   - Torpidinidae
Type   - Torpedo (Electric ray).

Habit and habitat. Torpedo or Astrape is a marine fish, found on flat, sandy or muddy bottom at a depth of 40-50 fathoms. It is carnivorous.
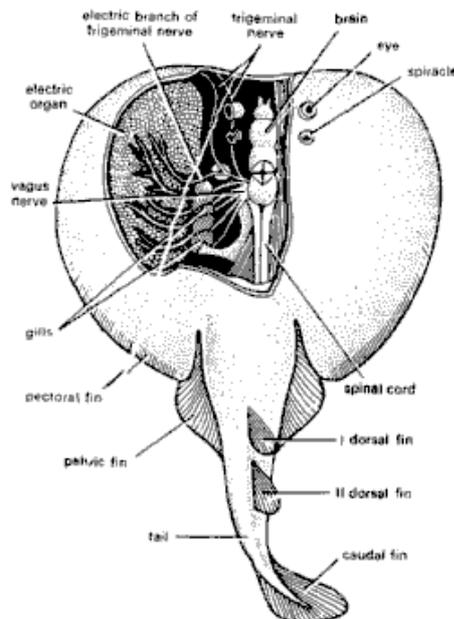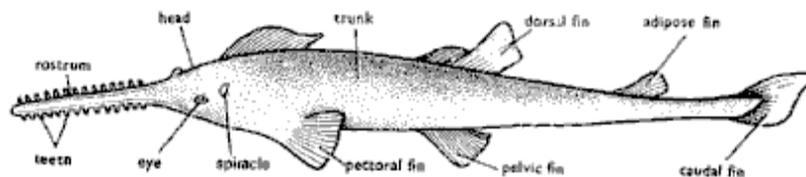
Comments:

(1) It is commonly known as Electric ray because of the presence of a pair of electric organs, one on either side of the body between head and the pectoral fins.

(2) Body is regionated into anterior semicircular disk supported by endoskeleton and posterior tail. Fish measures 60-90 cm across the widest part of the disk and the whole body has brown has brown background which is ornamented with beautiful irregularly shaped, magenta-coloured spiral and spots.

(3) Semicircular region is supported by branched prenasal rostrum and laterally by branched pre-orbital cartilages. Branches radiate towards periphery.

(4) Disk is bordered by pectoral fins.

(5) Skin is smooth, non-tuberculate and without scales.

(6) Eyes and spiracles are closely placed above electric organs dorsally.]

(7) Mouth is transverse and ventrally situated.

(8) Tail is thick and short with two dorsal fins, a caudal fin and two lateral folds of skin. Plevic fins are just beneath the lower margin of the pectoral fin.

(9) Gill-slits on the ventral side. Viviparous and produces live young's.

Identification. Since this fish has 2 building electric organs and above features, hence it is Torpedo.

iv). Pristis (Saw Fish)

Classification. Same as in Torpedo. Family Pristidae.

Habit and habitat. Pristis is a warm water marine type. It is predacious, feeding on small fishes and other marine animals by slashing them with its saw. It often ascends the river.
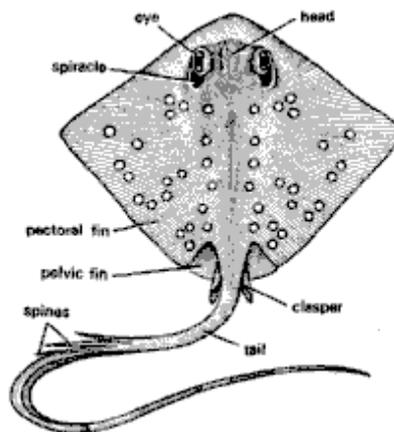

Comments:

(1) Pristis is commonly called as saw-fish, weighing 350-1200 lbs and measuring 3-6 metres in length.
(2) Elongated, shark like body is slightly depressed and divided into head, trunk and tail.
(3) Body shape is midway between a shark and a ray. The anterior part is flattened dorso-ventrally and is ray-like while the posterior part, for more than half, is shark-like. It exhibits close relationship with rays.
(4) Head contains a pair of eyes and a pair of spiracles behind the eyes.
(5) Snout is anteriorly produced into a saw-like rostrum with large and small weakly embedded teeth.

(6)   Mouth is well developed and ends in a heterocercal caudal fin.

(7)   Tail is well developed and ends in a heterocercal caudal fin.

(8)   Dorsal fins are large. First dorsal fin is opposite to pelvic fin. Viviparous.

V). Trygon (Sting Ray)

Classification. Same as that of Torpedo, family Rajidae.

Habit and habitat. Trygon is found lying quietly on the sea bottom. It occasionally swims to change the place in search of prey or moves in self-defence. It is carnivorous feeding on szamll fish, crustacceans and molluscs. It also shows adaptive or protective colouration to conceal itself from the enemies.

Comments:

(1) Trygon (= Dasyatis) is commonly called as sting ray or whip-tailed ray because of the presence of a serrated sting at the base of the tail.

(2) It consits of huge kite-shaped fleshy body and long whip-like tail. Head and body dorso-ventrally compressed.

(3) Outer anterior margin of pectorals continuous along side of head upto end of snout forming sub-rhombic disc-shaped body. Disc less than 1.3 times as broad as long.

(4) Pectoral fins being confluent with the sides of the head, their pre-axial endoskeleton radiates to meet in front of the skull along the lateral margins of prenasal rostral cartilage. Pelvic fins small.

(5) Skin is smooth or spiny.

(6) Mouth is ventral and rectangular. Nasofrontal flap is present in front of the mouth.

(7) Head contains a pair of dorsal eyes.

(8) Spiracles present behind the eyes.

(9) Gill-slits 5 paris, ventral in position.

Idenfication. Since this fish has ship-like tail and above features, hence it is Trygon.

vi) Amia (Bowfin)

Classification

Phylum	- Chordata
Group	- Craniata
Subphylum	- Vertebrata
Division	- Gnathostomata
Superclass	- Pisces
Class	- Osteichthyes
Superclass	- Actinoptergii
Superorder	- Holostei
Order	- Amiiformes
Family	- Amiidae
Type	- *Amia calva* (Bow-fin)

Habit and habitat. Amia is a freshwater and carnivorous fish, feeding voraciously upon other fishes, insects and crustaceans.
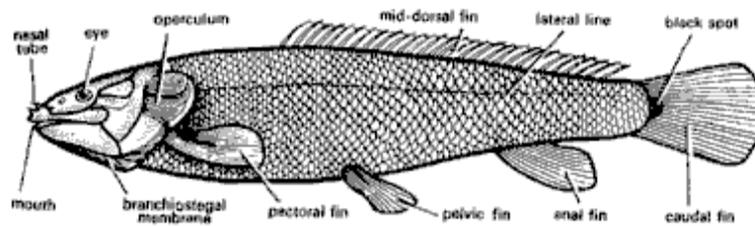
Comments:

(1)   Amia commonly known as bow-fin, measures about 60cm.
(2)   Shape of body is fusiform, compressed and divided into head, trunk and tail.
(3)   Head contains ventral mouth and dorsal eyes. The terminal mouth has a thick upper lip and a pair of dorsal barbells.
(4)   Mid dorsal fin has long base and continuous for the greater part of the trunk and tail.
      All fins are short and devoid of fulcra. Anal fin is situated near fan-shaped caudal fin.
(5)   Tail is homocercal.
(6)   Sexual dimorphic. Males are smaller than females and provided with a rounded spot at the base of caudal fin.

Idenfication. Since this fish has continuous dorsal fin and above features, hence it is *Amia*

vii) Notopterus (Chital)

Classification

Phylum	-	Chordata
Group	-	Craniata
Subphylum	-	vertebrata
Division	-	Gnathostomata
Superclass	-	Pisces
Subclass	-	Actinopterygill
Superorder	-	Teleostel
Order	-	Ostariophysi
Family	-	Notopteridae
Type	-	Notopterus

Habit and habitat. Notopterus, commonly inhabits marshy meadows, lakes, feshwater of brackish water. It is a bottom feeder, carnivorous, predacious and feeding on small organisms, insects and crustacceans.
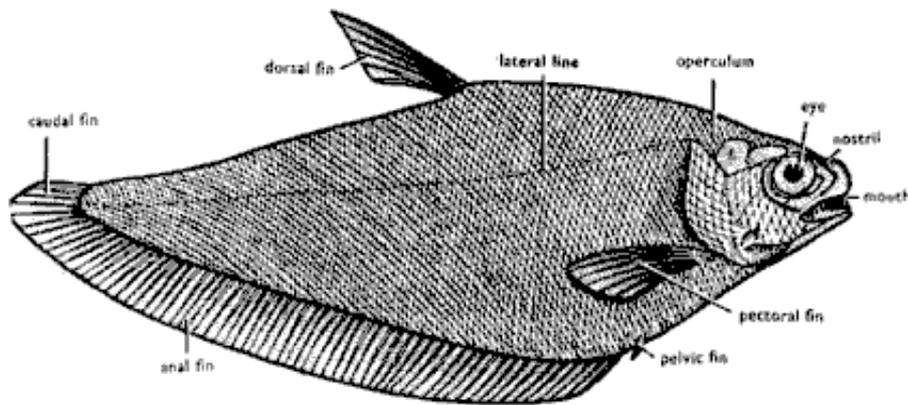
Comments:

(1) Notopterus is commonly known as cat-fish or chital.

(2) Body is very strongly compressed with a short pre-caudal region and measuring about 11/2 meters in length.

(3) Colour is silvery dark or greenish or glossy yellow on the back.

(4) Head contains large and oblique mouth, whitish eyes and nostrils. The musciferous channels well developed on the head.

(3) Dorsal fin is short and ventral fin very much reduced or absent. Very much elongated anal fin confluent with reduced caudal fin. Number of combined rays of anal and caudal fin

varies from 85-100. Pelvic fin has 3-6 rays. Sub-opercular absent.

(4)   Gill covers are scaly,. Lateral line scales 120-180, ventral scutes 25-45.

Identification. Since this fish has confluent anal and caudal fins, strongly compressed body and above features, hence it is Notopterus.

viii) Labeo rohita (Rohu)

Classification. Same as in Notopterus. Family Cyprinidae.

Habit and habitat. Leabeo rohita is abundantly found in ponds and rivers. Carps are vegetarian and bottom feeders. They can occasionally feed opn animal diet. Because of its feeding habit, it is cultivated with two other carps, Catla catla and Cirrhina mrigala. Rohu breeds only in the rivers and bund type of tanks but not in confined waters.
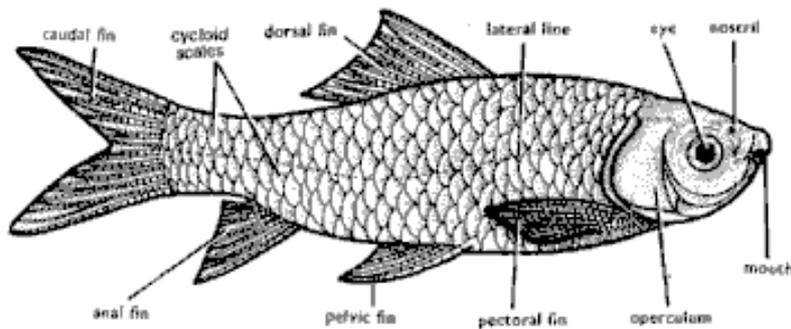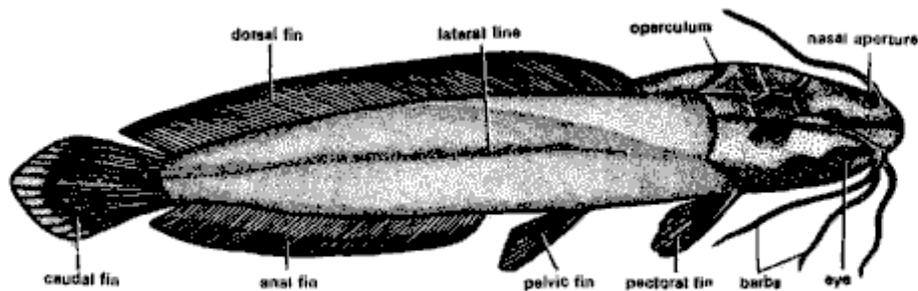
Comments:

(1)   Labeo rohita is commonly known as carp and Rohu in Hindi.

(2)   Body compressed, fusiform, about 1 metre in length and weighing about 4 kg.

(3)   Colour of the body is bluish or brownish on back an silvery white below. Body covered with large overlapping cycloid scales.

(4)   Body is regionated into head, trunk and tail.,

(5)   Head is depressed and is produced into a short, obtuse and blunt snout. It bears a subterminal fringe-lipped mouth bounded by fleshy upper and lower lips. It also contains paired nostrils and paired eyes.

(6)   A pair of filamentous barbels arises from upper lip. Small tubercles cover the snout, which is oblong, depressed, swollen and projecting beyond the jaws.

(7)   Large operculum hangs on either side enclosing gills and branchial chamber.

(8)   Lateral line is distinct. Scales overlying the lateral line are perforated by tubes of the lateral line system. Scales are of taxonomic value. Scales are flat, bony with rounded edges and are called as cycloid scales. These overlap and form a complete covering.

(9)   Labeo contains dorsal, anal, caudal, paired pectoral and pelvic fins. Fin rays are soft.

Idenfication: Since this fish has overlapping scales and above features, hence it is *Labeo rohita*

ix) Clarius (Magur)

Classification. Same as in Notopterus. Family Claridae.

Habit and habitat. Clarius batrachus is found in fresh and brackish waters. It takes a wide variety of food including clams, insect larvae and crustaceans living in dirty ponds and muddy water. They act as scavangers.

Comments:

(1) Clarius is commonly called as cat-fish or magur. Body is divided into head, trunk and tail.
(2) It is characterized by its spikeless dorsal fin, which extends all along the body; pectoral fin is inserted very low; anal fin is not confluent with caudal.
(3) Head is flat with four pairs of non-contractile and sensory barbels. Head bones are superficially exposed.
(4) Body is covered by scaleless and naked skin.

(5) Eyes reduced and spiracles absent.

(6) Tail is laterally compressed. Diphycercal and having rounded caudal fin.

(7) The pectoral fins cause painful wounds. They are place very low along ventro-lateral angles of abdomen.

Identification. Since this fish has peculiar dorsal fin, brabels and above features, hence it is Clarius.

x) Wallago attu (Lachi)

Classification. Same as in Notopterus.

Habit and habitat. Wallago is found in temperate and tropical fresh waters, inhabiting deep flowing waters of rivers and tanks in hilly and low country regions. It is predacious and feeds on young carps.
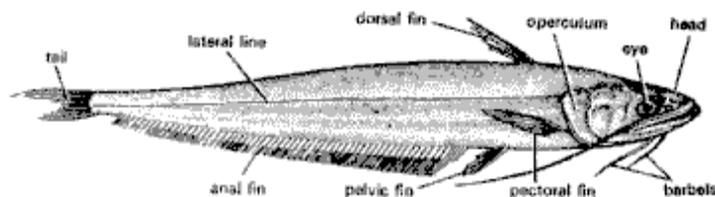
Comments:

(1) Wallago is also called as cat-fish or Lachi.

(2) Colour of the body varies. Dorsally it is greyish brown, head is purplish and belly whitish.

(3) Body is divided into head, trunk and tail. Head is very large, trunk small and tail long and tapering. Jaws provided with villiform teeth.

(4) Clefts of the mouth extend behind the orbits.

(5) Head contains nostrils, 2 maxillary and 2 mandibular sensory barbels.

(6) Eyes are found above the level of the mouth and not covered with skin.

(7) Dorsal fin is small like pectorals. It has fewer than seven rays and is not preceded by a spine. Adipose fin absent. Pectoral fin finely serrated. Anal fin large and confluent with the caudal fin. It is very much elongated and contains about 90 anal rays. Pelvic fins small.

(8) Scales are absent and body is covered with naked skin. Profile of the back oblique.

Identification.   Since this fish has smooth skin, small dorsal fin and above features, hence it is Wallago.
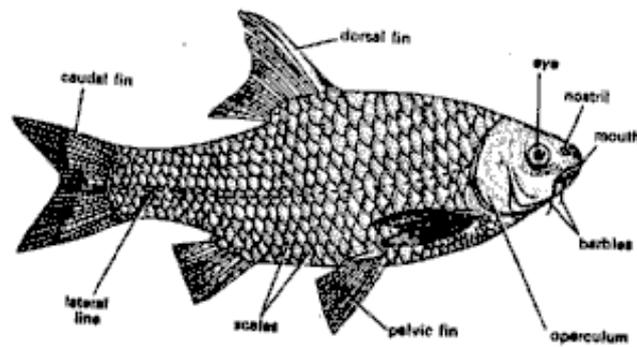
xi) Barbus sarana (Minnow)

Classification. Some as in Notopterus.

Family   -   Cyprinidae

Type   -   *Barbus sarana*

Habit and habitat. Barbus is a common freshwter fish of ponds, lakes and rivers.

Comments:

(1) Barbus is commonly called as Minnow or carp.

(2) Body of the fish is covered with large scales and is divided into head, trunk and tail.

(3) Head has an upward mouth without inner fold, small barbels, large eyes without adipose eye lid and devoid of scales.

(4) Lips thin without plicae or papillae; upper jaw bordered only by premaxillae.

(5) Fins are whitish or yellowish. Dorsal fin is opposite to anal fin and contains spine-like rays. Caudal fin rounded.

Identification. Since this ish has scaleless head, simple lips and ab ove features, hence it is Barbus.

xii)Ophiocephalus punctatus (Snake Head)

Classification. Same as in Notopterus.

order -   Ophicephaliformes

Habit and habitat. Ophiocephalus is commonly found in freshwater ponds and rarely in flowing waters. They are able to survive drought in semifluid or beneath dry mud and have an accessory branchial cavity for aerial breathing.
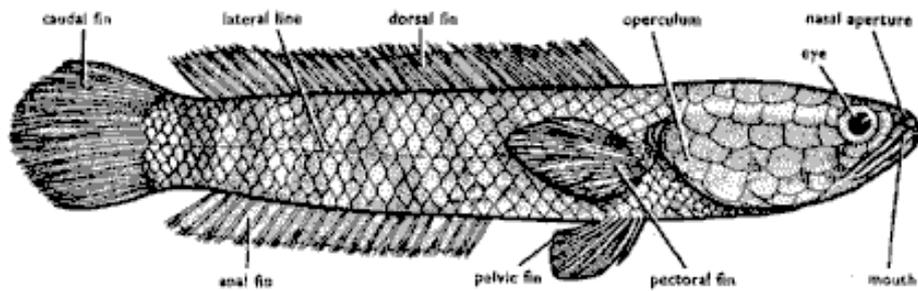
Comments:

(1)   Commonly referred to as snake-headed fish. Colour of the fish varies with water, with greeting back, yellowish sides and striped abdomen. Some specimens possess scattered dots on the head.

(2)   Body is elongated and cylindrical and differentiated into head, trunk and tail. Head and body covered with cycloid scales.

(3) Head triangular, tapers into a pointed snout. Teeth present on jaws and palate. Maxillae excluded from border of upper jaw. Lower jaw protruding beyond upper jaw.

(4) Dorsal and anal fins are long. Pectoral fins nearer to pelvic fins.

(5) Caudal fin is rounded.

(6) Lateral line is slightly curved.

Identification: Since this fish has characteristic anal and dorsal fin and above features, hence it is ***Ophiocephalus***

xiii) Anguilla(Eel)

**classification.** Same as in Notopterus.

Order - Apodes or Anguliformes
Type - Anguilla

Habit and habitat. Anuguilla is a freshwater fish. It is a voracious feeder and catadromous fish and it can live for several hours out of water. The adult eels live in ponds, estuaries, rivers and coastal areas of the sea and damp grass or moss outside water.

Comments:

(1) Anguilla is commonly known as eel, measuring 1.2 metres in length.

(2) Body is slender, elongated and snake like.
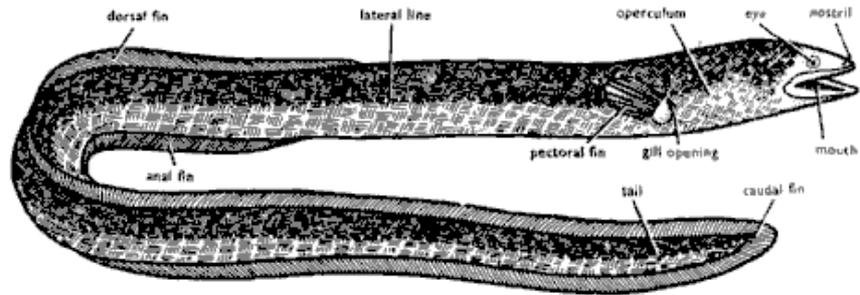
(3)   One each side operculum covers the gill slits.

(4)   Dorsal fin, anal fin and caudal fin are joined together forming a continuous fin. Pelvic fins are absent. Fins are supported by fin rays.

(5)   Body is covered by minuted scales embedded in the skin and arranged obliquely at right angles to one another forming a curious pattern.

(6)   Gills displaced posteriorly with 6-22 branchiostegal rays.

Identification: Since this fish has continuous caudal , anal and dorsal fins and above features, hence it is *Anguilla*.

xiv) Anabas (Climbing Perch)

Classification. Same as in Notopterus.

Order   -   Percomorphi
Family   -   Anabantidae
Type   -   Anabas (The Clibing Perch)

Habit and habitat. Anabas is a common South Indian freshwater fish. It can live out of water for a long period.
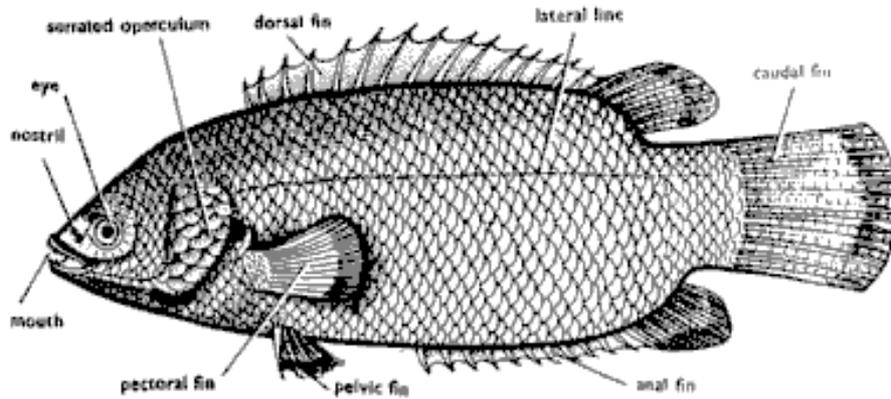
Comments.:

(1)   Anabas is commonly known as climbing perch.

(2)   Fish measuring about 30 cm is olive green in colour.

(3)   Body of the fish covered by cycloid scales and divided into head, trunk and tail.

(4)   Head is conical containing large eyes, nostrils and mouth. In front of eyes is a pre-orbital bone containing spines.Small spines also occur along the edge of operculum.

(5)   Dorsal and anal fins are elongated. They are divided into anterior and posterior parts, supported by stiff spines and soft rays respectively.

(6)   Pelvic fins are anteriorly situated almost below pectorals.

(7)   Tail is perfectly symmetrical.

Identification. Since this fish has dorsal and anal fins rays and above features, hence it is anabas.
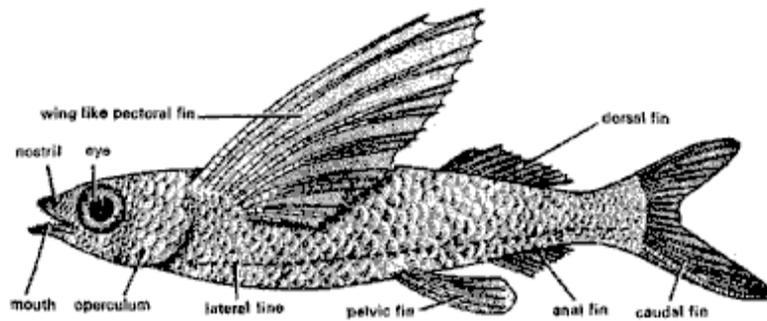
xv) Exocoetus (Flying Fish)

Classification. Same as in Notopterus.

Order -   Synentognathi
Type   -   Exocoetus.

Habit and habitat: Exocetus is found in sea, often skittering near the boats. It is pelagic and feeding on prawns and young fishes and their eggs. Small fishes live in sandy shoal-places near the coast.

Comments:

(1)   Commonly known as flying fish.
(2)   Elongated body and divided into head, trunk and tail.
(3)   Head contains large eyes. The upper part of the snout is produced into a process and contains nostril.
(4)   Mouth opening is small but teeth in both jaws.

(5) Dorsal and anal fins are short and oipposite to each pelvic fin.

(6) Pectoral fins are exceptionally large, spread like wings and make gliding flight.

(7) Ventral fin is well developed. Tail is hypoblastic. Oviparus.

Identification. Since this fish has large pectoral fins and the above features, hence it is ***Exocetus***.

xvi) Hippocampus (Sea Horse)
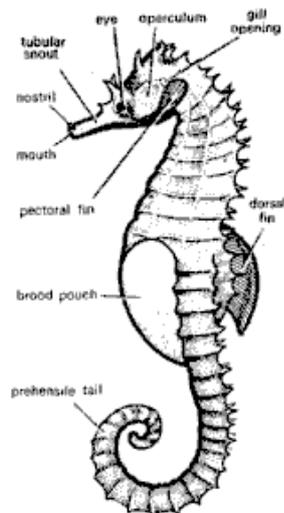
Classification : Same as in Notopterus.

Order   -   Solenichthyes
Family   -   Syngnathidae
Type   -   Hippocampus.

Habit and habitat. They swim upright swaying their tails and gyrating their trunks in graceful manner, holding a weed with their tails.

Comments.

(1) Hippocampus is commonly called as sea-horse because its anterior end is shaped like the neck and head of a horse.

(2) Body is divided into head, trunk and tail., size varies from 5 to 17 cm.

(3) Head is produced anteriorly into snout and backwardly into a crest. Mouth is found at the end of snout. Eyes are situated on the upper side of head. There are two super orbital spines directed backwards and outwards.

(4) Some species have fine trailing filaments over the body.

(5) Body is covered by a rigid exoskeletal armour of ring-like bony plates.

(6) Dorsal fin is single, ventral and cudal fins are absent.

(7) A small transperant pectoral fin is found on either side of the head.

(8) Females have a small anal fin. Males contain brood pouch.

(9) Tail is prehensile.

Idenfication.: Since this fish has horse shaped head and snout and above feature, hence it is ***Hippocampus***.

xvii) Mystus seenghala

Class   -   Pisces
Sub-class   -   Teleostomi
Order   -   Cypriniformes
Family   -   Bagridae
Type   -   Mystus seenghala.

Habit and Habitat: It is a fresh water species found in India, Pakistan, Burma, Srilanka. It is a predatory in nature and highly carnivorous. It is a coloum and selective feeder. This fish consumes other fishes, insects, prawns, crabs, mollusks and amphibians.

Comments:

1   Body is elongated, brownish above with silvery sides.
2   It has a moderate adipose fin with a distinguishing feature of a circular black spot on the adipose fin.
3   Four pairs of barbles are present, maxillary barbell elongated, extended beyond the pelvis, mandibular barbell is small.

4	A deeply forked caudal fin with the upper lobe longer than the lower.

5.	The snout is distinctly longer withlarge upper jaw.

xviii) Ompak bimaculatus (Butter fish)

Class	-	Pisces

Sub-class	-	Teleostomi

Order	-	Cypriniformes

Family	-	Siluridae

Type	-	Ompak bimaculatus

Habit and Habitat: This is found in fresh water of India, Pakistan, Srilanka, Burma and Indo-China. It is a carnivorous, surface and selective feeder. The main food of the fish is small and medium sized fishes, insects and their larvae.
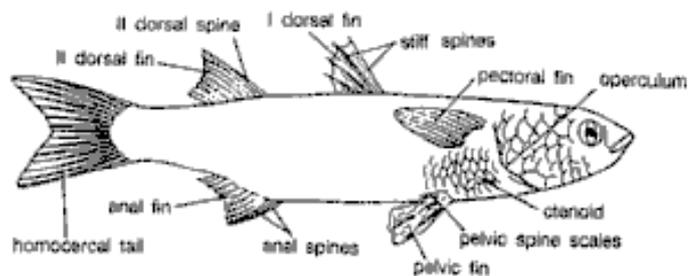
Comments:

1.	Body is elongated , laterally compressed , silvery or grayish brown in colour.

2.	Body is strongly compressed and the head is depressed with an oblique place mouth with a prominent lower jaw.

3. Dorsal fin is short or absent. The adipose fin is absent. The anal fin is moderately long.
4. Two pairs of barbells, maxillary pair reach the origin of the anal fin or even beyond and the mandibular pair reach the hinder edge of the orbit.
5. A black spot on the shoulder and another spot at the tail is present.
6. Caudal fin is deeply forked with sharp lobes.

xix) Mugil Cephalus(Grey stripped mullet)

Class   -   Pisces
Sub-class   -   Teleostomi
Order   -   Mugiliformes
Family   -   Mugilidae
Type   -   Mugil cephalus.

Habit and Habitat: Marine and esturine fish suitable for cultivation in fresh and brackish water ponds. It is an filter feeder. It is an omnivorous, planktonphagous feeds on small crustaceans.
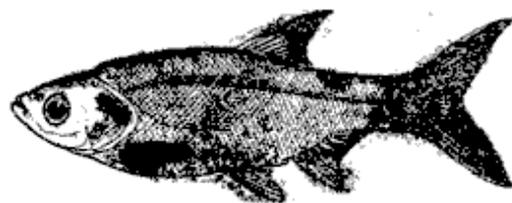
Comments:

1. Body is elongated, slightly compressed from side to side, with dull brown on dorsal surface and lighter along the abdomen. Dorsal and caudal fins are grayish.
2. Head is short and flattened with broad terminal mouth. Upper lip is thin and smooth.
3. Two dorsal fins, the first is short with 4 slender spines, arises midway between the end of the base of the caudal fin and snout. The second dorsal fin is usually with 8 soft rays.
4. Pectorals are situated above the middle of the depth of the body. Anal fin originates opposite to the second dorsal fin.
5. Caudal fin is with pointed lobes.
6. A dark black spot on the pectoral base is present.

xx) Amblypharyngodon mola

Class   -   Pisces
Sub-class   -   Teleostomi
Order   -   Cypriniformes
Family   -   Cyprinidae
Type   -   Chanos chanos

Comments

1.   Rounded Abdomen. Dorsal body profile is slightly more convex than the Abdominal one.
2.   Head is compressed and snout is thin .
3.   Upper lip is absent mouth with Prominent lower jaw and elongated.
4.   Eyes are divoid of adipose membrane and are locate in middle of the head.
5.   Dorsal fin with concave upper edge, originates behind ventral fin and it is longer.

xxi) Chanos chanos(Milk fish)

Class   -   Pisces
Sub-class   -   Teleostomi
Order   -   Clupeiformes
Family   -   Chanidae
Type   -   Chanos chanos

Habit and habitat: It tolerate a wide range of fluctuations insalinity. Marine and esturine fishes are suitable for fresh and brackish water ponds. It is primarily a phytoplankton feeder.

Comments:

1.   Body is compressed, elongated, beautifully shaped with silvery green along back. Body depth is shorter than or equal to that of its head length.
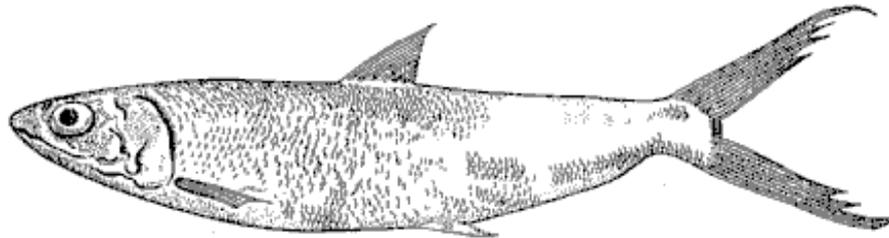
2. Mouth is small, terminal and transverse. Upper jaw over hanging the lower jaw.
3. Single dorsal fin with 13-16 rays and arises mid way between the front edge of the eye and the base of caudal.
4. Caudal fin is forked with sub-equal lobes.
5. Ventral fin is inserted under the middle of the dorsal fin.

xxii) Macrobrachium rosenbergii (Gaint fresh water prawn)

Phyllum   -   Arthropoda
Class   -   Crustacea
Sub Class   -   Malacostraca
Order   -   Decapoda
Family   -   Palaemonidae

Habit and Habitat: It occurs in rivers, estuaries and coastal areas. It migrates to estuaries during the breeding season. It is an omnivorous bottom feeder, feed up on mollusks, worms, insects, small crustaceans, vegetable matter and stems of aquatic weeds.
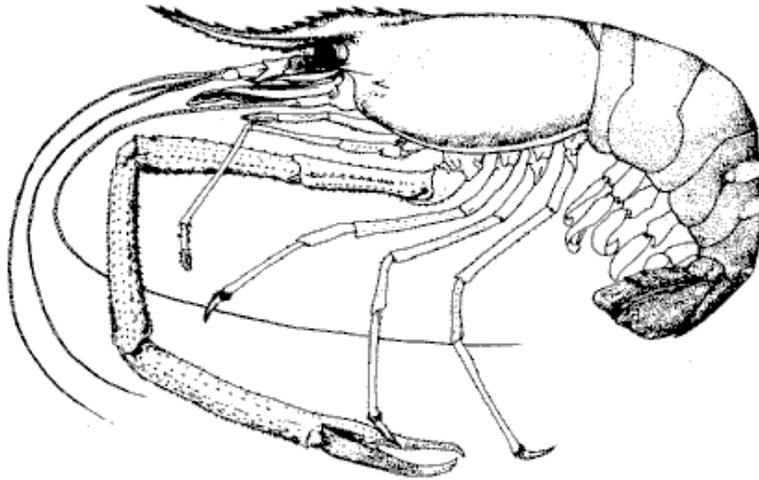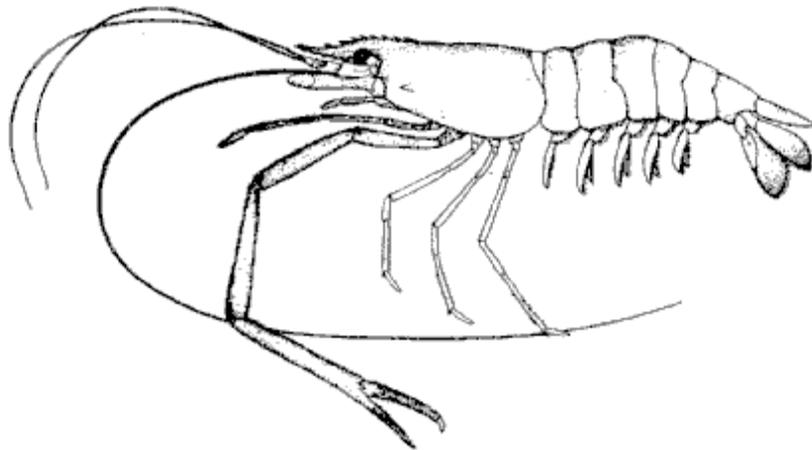
Comments:

1. Body is divisable into anterior rigid cephalo-thorax, posterior flexible abdomen and a post segmental terminal telson. All these divisions contains appendages

2. Rostrum long and slender, usually extending and bears 13-14 rostral spines dorsally and 11 rostral spines ventrally.

3. The second pair of walking leg of male develops abnormally with well developed chela and thus sexual dimorphism is exhibited. The legs are stout.

4. Telson regularly tapering to a sharp point, without a posterior margin, tip over reaching postero;ateral spines.

5. No pubescence on the rest of the legs, except for scattered hairs.

6. Body colour is dark grey, some times with longitudinal or irregular streaks of dark and lighter colour; often orange patches at the articulation of abdominal somites.

xxiii) Macrobrachium molcolmsonii (River Prawn).

Classification as above

Habit and Habitat: It occurs in rivers and migrates from rivers to estuaries during the breeding season. It is a bentophagic omnivore and feeds on worms, insects, mollusks, crustaceans, vegetable matter, algae and weeds.

Comments:

1. Body is divisable into anterior rigid cephalo-thorax, posterior flexible abdomen and a post segmental terminal telson. All these divisions contains appendages
2. Rostrum is short and straight. It bears 11-12 dorsal spines and 4-5 ventral spines.
3. The second pair of walking legs of males develops chela and exhibit the sexual dimorphism. The second pair of legs are longer than the body and are stout.

4. Telson gradually tapering to a sharp point, without posterior margin.

5. The body and anterior two pairs of peropods dark grey, paler ventrally; antennae and inner flagella of antennules grey; outer flagella brownish.

xxiv) Penaeus monodon (Tiger Prawn)

Phyllum	-	Arthropoda
Class	-	Crustacea
Sub Class	-	Malacostraca
Order	-	Decapoda
Family	-	Penaeidae

Habit and Habitat: It is a marine and deep sea form, chiefly inhabiting in tropics. It feeds on both animal and plant materials. It is a detritivore.
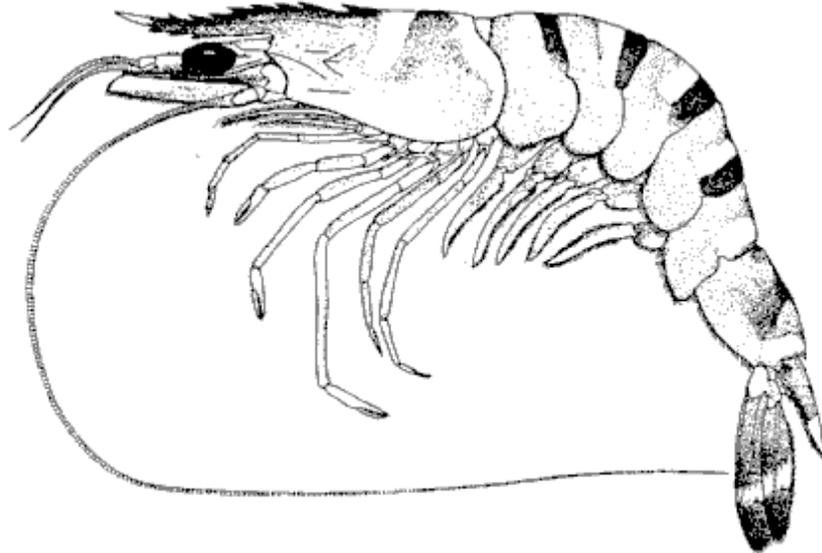
Comments:

1. Body is divisable into anterior rigid cephalo-thorax, posterior flexible abdomen and a post segmental terminal telson. All these divisions contains appendages.
2. Cephalothorax is formed from 6 cephalic and 8 thoracic segments. Head contains two prominent eyes, very long antennae and short antennules.
3. Abdomen consists of 6 segments and laterally compressed but not bent sharply.
4. Rostrum strongly sigmoidal with the rostral formula 7-8 teeth on dorsal and 2-3 on ventral margins.
5. Pleopods are brown to blue with distinct yellow bands and exopodite is absent on fifth pleopod.
6. Body is dark brown or reddish in colour with transverse bands.Telson consist of two lateral lobes closely meeting along the median line.

xxv) Penaeus indicus (White Prawn).

Habit and Habitat: This is confined to East and West coasts. It feeds on detritus, small crustaceans, molluscnas, polychetes.
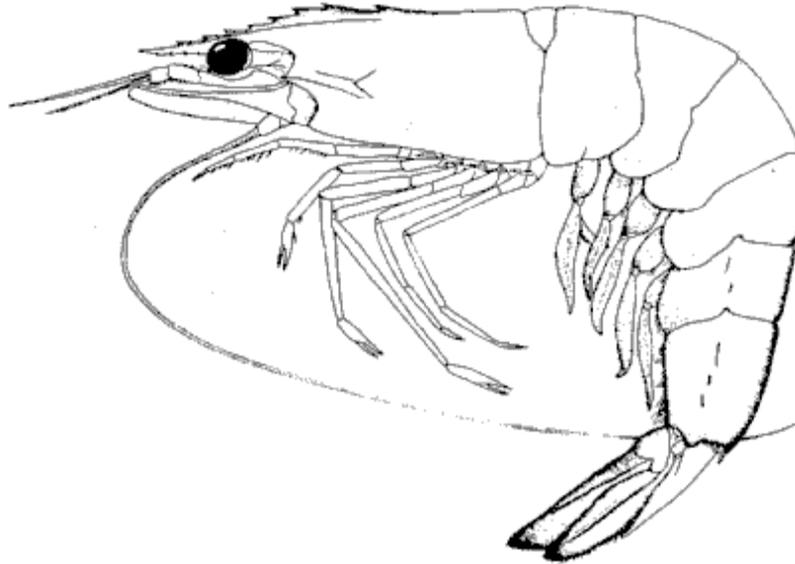
Comments:

1. Body is divisable into anterior rigid cephalo-thorax, posterior flexible abdomen and a post segmental terminal telson. All these divisions contains appendages.
2. Cephalothorax is formed from 6 cephalic and 8 thoracic segments. Head contains two prominent eyes, very long antennae and short antennules.

3. Abdomen consists of 6 segments and laterally compressed but not bent sharply.
4. This species shows large variations in the rostral length. In young forms, it is slender, elongated and surpasses the tip of the antennal scale. In adult, the length is reduced and thickened at the base. It bears 7 to 9 teeth on dorsal and 4-6 teeth on ventral margin.
5. In adult male, dactlylus of third maxillipede is as long as propodus.
6. Body is pale pink to yellowish, semi-translucent, with olive green to grey blue speckles.

2. Identification of Scales :

(i). Fish : Cycloid Scales.

Procedure: Take out one or two scales from any bony fish, e.g. Labeo. Wash the material with the water, dehydrate it and stain with borax – caramine. Clear in xylol and mount it on a slide and study under microscope.

concentric lines of growth
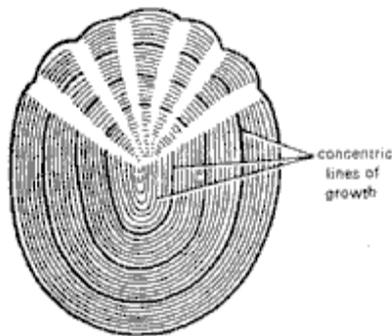
Comments:

(1)   The cycloid scales are found embedded in dermis and are almost spherical in outline.

(2)   Its outer surface is smooth and circular lines are found on it. Some of these lines are thick and can determine the age of fish. These are the line growth..

(3)   The posterior part of each scale overlaps the anterior part of the scale behind.

(4)   Cycloid scales are found in many bony fishes, dipnoi or some osteolepids..

(ii). Fish: Placoid Scales

Procedure: Cut a small piece of skin from the back of the scoliodon. Place the skin in watch glass full of water and then remove all muscles from the ventral surface by erasing with a blade. Continue this process till the skin becomes transperant. Take this piece of skin, place it in a test tube with 1% KOH solution. Fill the test tube to half with the water. Boil the material on a spirit lamp for 2 or 3 minutes. Shake the test tube and the KOH solution continuously. Place a drop of this solution on to the slide and observe under the microscope.
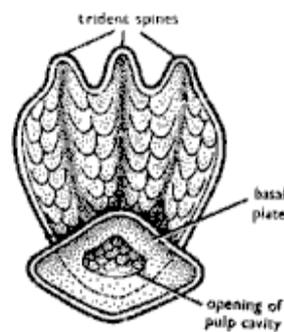
Comments :

(1)   With very few exceptions, placoid scales are abundantly found in dermis of elasmobranch fishes.

(2)   Placoid scales are arranged in regular oblique rows. They are dermal in origin and cover entire surface of the body, forming dermal exoskeleton of the sharks.

(3)   Each scale is composed of a basal bony plate embedded in the dermis, from which a spine projects upwards and points posteriorly.

(4) Basal plate is formed of a trabecular calcified tissue.

(5) Spine is composed of dentine covered by a hard material, vitrodentine.

(6) Placoid scale contains a pulp cavity in spine.

(7) Pulp cavity contains odontoblasts, dentine forming cells, blood capillaries, nerves and lymph channels.

(8) General similarity in structure of placoid scales to teeth of higher forms should be aparent. Both are considered to be remnants of bony armour of such primitive vertebrates as ostracoderms and certain placoderms.

Identification. Since this scale has trident spines, hence it is palcoid scale.

(iii). Fish: Ganoid Scales
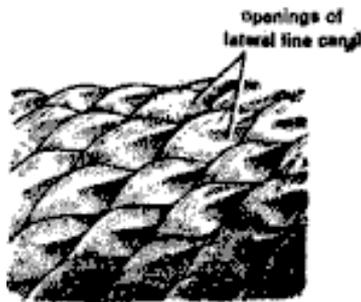
Comments:

(1) Ganoid scales are found in primitive ray-finned fishes such as Ploypterus and gar pikes.

(2) Scales are covered with a hard, shiny and translucent material of mesodermal origin called as ganoin.

(3) Ganoid scales fit together like tiles and are arranged in diagonal rows.

(4) Scales are dermal in origin.Each scale consists of a bony base, coated by shining substance called as ganoin.

Identification. Since the above scale is overlapping & fitted like tiles, hence it is ganoid scale.

(iv).Fish: Ctenoid Scales.

Procedure: Take out one or two scales from any teleost like Anabas. Wash the material with the water, dehydrate it. Mount it on to the slide and study under the microscope.

concentric lines of growth

tiny teeth
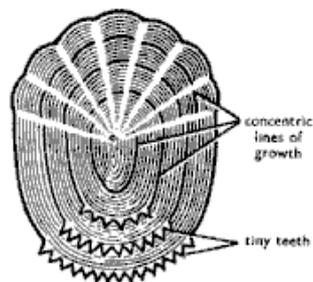
Comments:

(1) Ctenoid scale is horny plate like structure with an elobarate series of tooth like process at the free edge, giving it a comb like appearance.

(2) There are lines of growth present over the scales and can determine the age of the fish.

(3) The posterior part of each scale overlaps the anterior part of the scale behind.

3.   Dissections of fish and prawn:

(i). Fish – Digestive system:

Procedure: Pin down the fish through the fins in dissecting tray. Locate the positions of the pectoral and pelvic girdles by tracing them through the skin. Now give a mid-ventral incision extending from pelvic girdle to the pectoral girdle. Cut open the body cavity, wash it and observe the following structures:

Alimentary canal: It is an elongated tube from mouth to anus. It is differentiated into various parts such as mouth, buccal cavity, pharynx, oesophagus, stomach, intestine and rectum.

(a). *Mouth* – It is obtained by upper and lower lips and opens into the buccal cavity.
(b). *Buccal cavity* – It is a darso- ventrally depressed chamber. A distinct tongue is lacking.
(c). *Pharynx* – It is also darso-ventrally flattened chamber. It is divisible into an anterior pharynx, being perforated on the sides by gill slits and posterior pharynx being provided with pharyngeal teeth.
(d). *Oesophagus* – It is a narrow, short but thick tube.
(e). *Stomach* – It is thick walled tube and can be differentiated into the anterior cardic region and the posterior pyloric region.
(f). *Intestine* – It is very much coiled and a thin walled tube of more or less uniform diameter.
(g). *Rectum* – It is also a thin walled chamber of more diameter than intestine and opens to the exterior by anus.
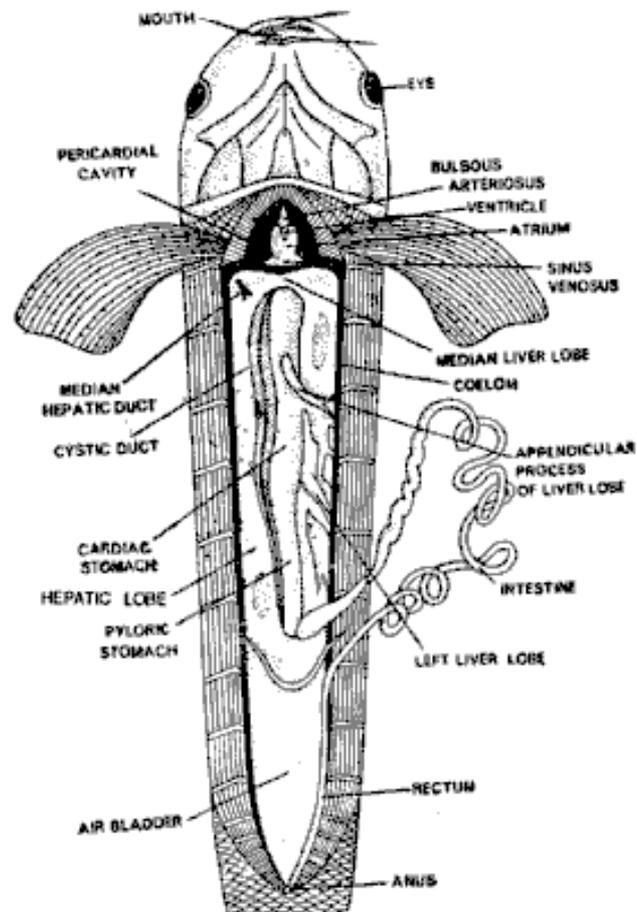
Associated digestive glands:

(a). *Liver* – It is an elongated and solid bilobed gland of dark brown colour. The right lobe is long but narrow while the left lobe is broader. The two lobes are connected at different places. The gall blader is an elongated thin walled
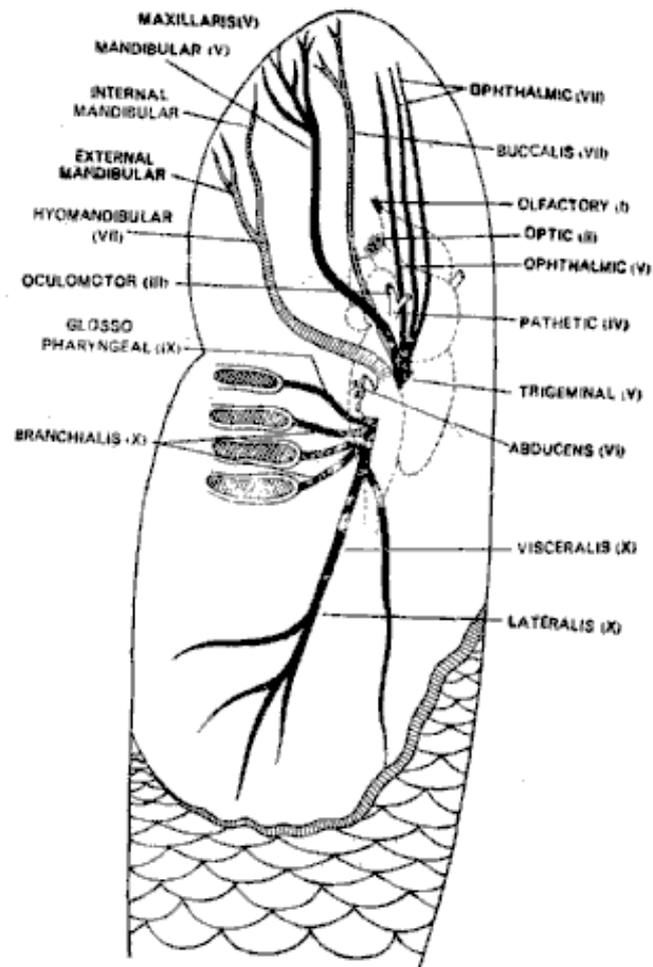
sac, lying between the right lobe of liver and the cardiac part of the stomach.

(b). *Pancreas* – A conspicuous or apparent gland is absent in Labeo, but it is actually present in a difused from scattered more or less all over the visceral cavity and partly being embedded in the liver and spleen.

ii). Fish – Nervous system:

Procedure: Remove the skin of the head completely and expose the brain by scraping the bone from the dorsal side of the brain. Remove the eye ball and bones surrounding it. Immediately dorsal to the orbit lie two slender nerves. Dissect them behind the place of their origin. Observe the following cranial nerves.

Cranial nerve	*Name of the nerve and its description*
I	*Olfactory nerve*- arise from the olfactory cells in the olfactory organ and directly passes into the lobes of the brain.
II	*Optic nerve* – arise from the optic lobe and innervates in to the retina.
III	*Oculomotor nerve* – arise from the ventral side of the brain below the optic lobes, runs outwards and enters the orbit.
IV	*Trochlear nerve* – arises from the dorsal side of the brain between the optic lobe and the cerebellum.
V	*Trigeminal nerve* – divides into three branches.

MAXILLARIS(V)

MANDIBULAR (V)

INTERNAL MANDIBULAR

EXTERNAL MANDIBULAR

HYOMANDIBULAR (VII)

OCULOMOTOR (III)

GLOSSO PHARYNGEAL (IX)

BRANCHIALIS (X)

OPHTHALMIC (VII)

BUCCALIS (VII)

OLFACTORY (I)

OPTIC (II)

OPHTHALMIC (V)

PATHETIC (IV)

TRIGEMINAL (V)

ABDUCENS (VI)

VISCERALIS (X)

LATERALIS (X)

i). Opthalmic – arise from the anterior roof of the nerve and runs forward in close association of the opthalmicus superficialis.
ii). Maxillary – runs along the posterior margin of the orbit and turns towards the muscles of that region.

iii). Mandibular – It is the posterior part of the maxillary which cross the angle of the jaws and innervates the masticatory muscle, gum and teeth of the lower jaw.

VI   *Abducens nerve* – small nerve, innervates to the eye Muscle

VII   *Facial nerve* - arises just behind the fifth nerve and divides into three branches.
i). Opthalmicus superficialis – arises from the anterior root and run forward in close association of the opthalmicus of the fifth cranial nerve and innervates the skin of the dorsal surface of the snout.
ii). Buccalis – after its origin, it runs obliquely downwards and forwards across the orbit of its origin and innervates the associate regions.
iii). Hyomandibular – it runs backwards and outwards and divides into a mandibular externus, a mandibular internus and a hyoidean.

	iv). Palatinus – it runs across the floor of the orbit to the palate.
VIII	*Auditory nerve* – small nerve which arises directly from the medulla oblongata and passes to the auditory capsule.
IX	*Glossopharyngeal nerve* – arises from the ventral side of the medulla oblongata and innervates into first gill and divides into pre-trematic and post- trematic.
X	*Vagus nerve* – on emerging divides into three branches.

    i). Branchialis – innervates into all the gills except first.

    ii). Visceralis – it is the main branch which enters the body cavity and innervates the various visceral organs.

    iii). Lateralis – it runs backwards and runs parallel to the lateral line.

iii). Fish – Weberian Ossicle:

Procedure: Remove the skin and muscles of the region posterior to the operculum. Race out the air bladder and see attached to it anteriorly a triangular piece of bone known as weberian ossicle. Carefully trace it further anteriorly and take it out and place the material in watch glass.
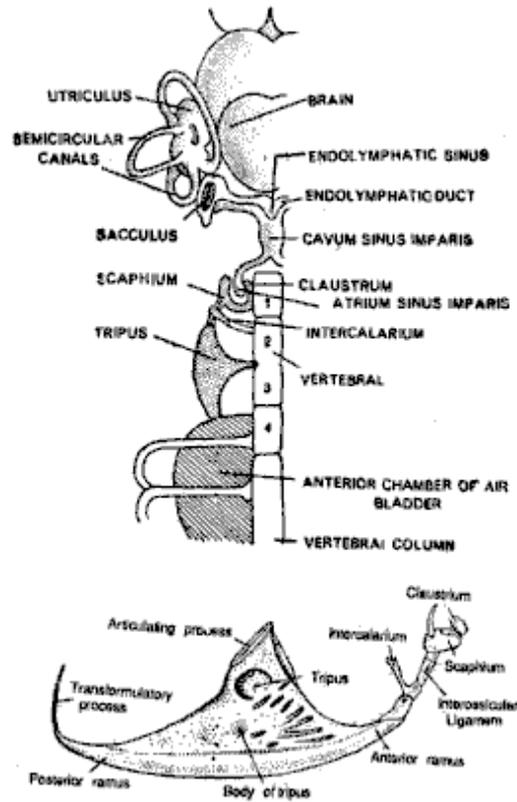
Structure:

1. The first bone is attached to the internal ear and is known as claustrum.

2. This is followed by other bones in chain called the Scaphium, the Interclarium and the Tripus.

3. The tripus is the largest bone of the series attached to the air bladder posteriorly.

4. It is suggested that sound waves travel to the internal ear through this chain of bones.

iv). Fish – Pituitary gland :

Fish pituitary gland is small, soft body and creamish white in colour. It is more or less in round in carps. It lies on the ventral side of the brain behind the optic chiasma in a concavity of the floor of the brain box, known as Sell turcica and enclosed by a thin membrane called duramater. In few fishes, it is attached to the brain by a thin stalk, known as the infundibular stalk. The size and weight of the gland varies according to the size and the weight of the fish. In *Labeo rohita*, the average weight of the pituitary gland is 6.6 mg in 1-2 Kg fish, 10.3 mg in 2-3 Kg fish, 15.2 mg in 3-4 Kg fish and 18.6 mg in 4-5 Kg fish.

The fish donating the pituitary gland is called the donor fish. There are several techniques adopted for the collection of the gland. The most commonly adopted technique of gland collection is by chopping off the scalp of the fish skull by an oblique stroke of a butcher's knife. After the scalp is removed, the grey matter and fatty substances lying over the brain are gently cleaned with a piece of cotton. The brain thus exposed is carefully lifted out by detaching it from the nerves. In most of the cyprinids, when the brain is lifted, the gland is left behind on the floor of the brain box. The duramater covering the gland is then cautiously removed using a fine needle and forceps. The exposed gland is then picked up intact without causing any damage.

The gland after collection is immediately put in absolute alcohol and store at room temperature.

v). Prawn – Digestive system:

Procedure:

Take the specimen in hand or dish and lift the carapace from lateral side. Cut loose the carapace at its anterior margin to remove it completely. Remove all the abdominal terga and pleura with scalpel and forceps and expose the abdominal muscles. Pin the specimen in dissecting tray and remove completely the gonads to expose the digestive system. The stomach is found just beneath carapace, embedded in large digestive gland. The intestine is narrow tube present in the groove of abdominal muscles. For hastate plate or gastric mill, cut the stomach at both the ends and make a dorsal incision on the dorsal side. Spread it flat, clean, wash and then see its teeth and comb with the lens.

The digestive system is well developed with associated glands and is divided into three regions, Fore gut, Mid gut and Hind gut.

The fore gut consists of the following parts:
(a).   *Mouth* – It is a large aperture on the ventral side of the head bounded by labrum, mandible and labium.
(b).   *Buccal cavity* – Mouth leads into a short buccal cavity.
(c).   *Oesophagus* – It is a short duct arising from buccal cavity and it communicates with the stomach. It extendes upwards.
(d).   *Stomach*- It is a wide chamber consisting of cardiac and pyloric parts. Ventrally stomach is surrounded by orange – red hepatopancreas.
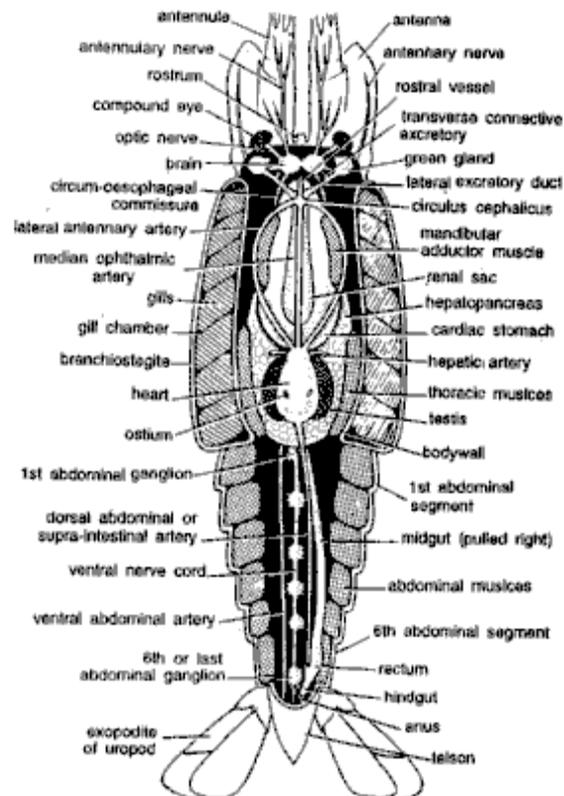
The mid gut is a very short and slender duct, which ascends between hepato-pancreasto extend backward.

The hind gut extends from mid gut to anus. The terminal part is modified as rectum.

vi). Prawn – Nervous system:

Procedure:

For nervous system first expose the nerve cord in the abdominal region. For this, cut medianly with scalpel between the large flexor muscles. Press these muscles and pin them in the dissecting tray. As the muscles are stretched, the nerve cord is very clearly seen with ganglionic swellings. Proceed from posterior side, cutting the middle line with the scissors through the chitinous endophragmal skeletal plates found on the ventral side of the thorax and exposing underlying nerves till brain is exposed. Study the various parts.

(a). *Brain or supra- oesophageal ganglia* – It is a bilobed ganglion, situated at the base of the rostrum. On each side, brain gives antennulary nerve to antennule, optic nerve to compound eye, statocystic nerve to statocyst, antennary nerve to antenna and tegumental nerve to labrum.

(b). *Circum-oesophageal commissures* – Brain give rise to a pair of thick posterior circum –oesophageal commissures, which surrounds oesophagus and unite together ventrally with sub-oesophageal ganglion. A transverse connective connects the two commisures. The sub-oesophageal ganglion is fused with anterior part of ventral thoracic ganglionic mass.
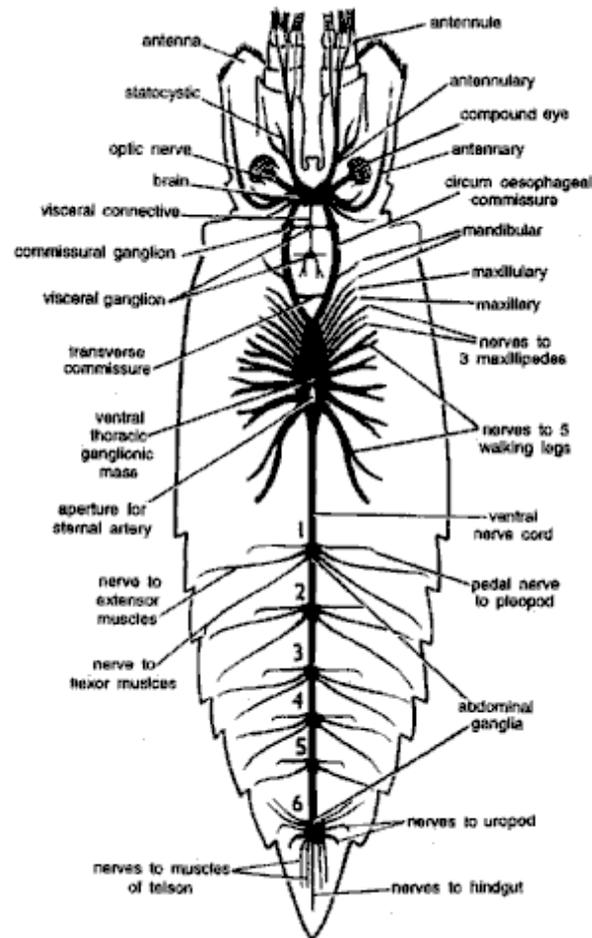
(c). *The ventral thoracic ganglionic mass* – It is formed by the fusion of eleven segment a caphalothoracic ganglia, which consequently gives 11 pairs of nerves.

(d). *The ventral nerve cord* – It runs in the abdomen, forming 6 ganglionic masses, from where nerves are given out to pleopods, musculature and uropod.

4. Identification of appendages of prawn:

For identification of the appendages in the preserved specimen- prawn, it should be placed in a dissecting tray with its ventral side facing upwards and the appendages of one side should be removed one by one. Starting with the posterior end, the abdominal appendages should be removed first, then the thoracic and lastly, the cephalic appendages. The base of each appendage should be held carefully with a pair of forceps and a circular incision made around the arthroidal membrane either with a pair of scissors or with an arrow-head needle. Each appendage should be carefully removed and arrange one in front of the other following the order from the cephalic, thoracic and abdominal appendages. They should be examined with the help of the description and diagram in the text given below. Care should be taken in removing the antennule so as not to injure the statocyst in it's precoxa.

There are 19 pairs of appendages in prawn, which are given out one on either side from each segment of the body. According to the division of the body into two main parts, the appendages are differentiated in to two categories. They are (i). **The cephalothoracic appendages** and (ii). **the abdominal appendages**, which are further demarcated into *cephalic* and *thoracic appendages*.

(1). ANTENNULE:

- Paired cephalic appendages which are situated beneath the eye-stalk one on either side.
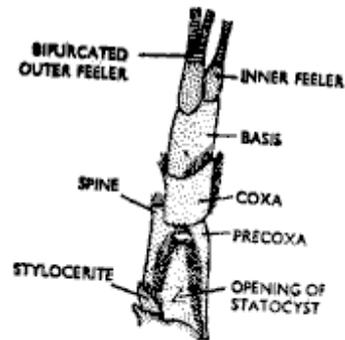
-   The protopodite is composed of precoxa, coax and basis.
-   The pre-coxa bears a small white, bead-like statocyst which is sensory in nature.
-   The coax is small cylindrical and is fringed with minute setae.
-   The basis possesses two many jointed and long feelers – the outer and inner.
-   The antennule is tactile in function.

(2). ANTENNA:

-   Paired cephalic appendages which are lying below each antennule.
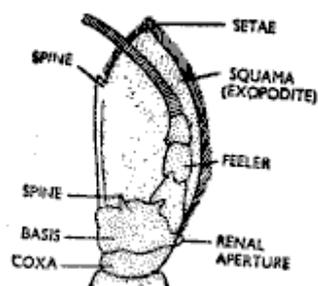
- The protopodite is composed of coaxa and basis.
- The coax is small ring like and bears the renal opening.
- The basis possesses a many jointed endopodite and a large and membranous squama, which helps in swimming.
- The squama is the modified exopodite.
- Antenna is excretory, sensory and balancing in function.

(3). MANDIBLE:
- Paired, stout and small cephalic appendages which lie on either sides of mouth.
- The coax is well developed and consists of a proximal apophysis and distal head.
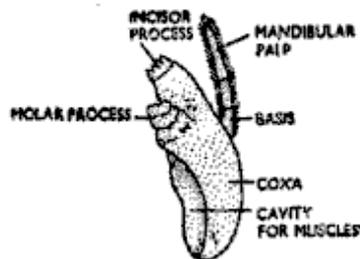
- The head bears a set of molar and incisor teeth each, which assist in mastication of food.
- The basis is many jointed and is known as mandibular palp.
- Exopodite wanting.
- It helps in mastication.

(4).MAXILLULA:

- Paired, leaf-like, small cephalic appendage which is situated beneath the mandible on either side.
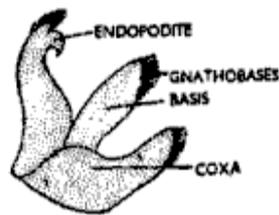
- The coax and basis bear the teeth like projections on its inner margin, which are called the gnathobases.
- The maxillula assists in the manipulation of food.

(5). MAXILLA:

- These are paired, leaf like appendages lying beneath each maxillula.
- The coax is small ring likeand divided partially.
- The basis forms the forked gnathobase on its inner margin.
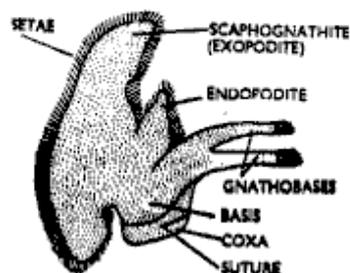
- Endopodite is small leaf like outgrowth, which is fringed with tactile setae.
- The exopodite is large, thin and cresentric shaped structure called Scaphognathite.
- The maxilla assists in respiration and the manipulation of food.

(6). FIRST MAXILLIPEDE:

- Paired, leaf like thoracic appendage lying just below each maxilla.
- The coxa and basis forms the gnathobase on its inner margin. The coxa bears a pair of primitive gill.
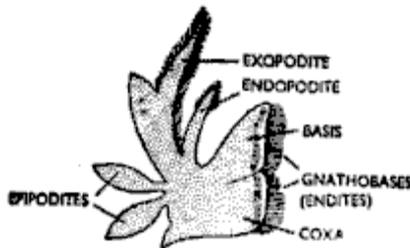
- The exopodite is long, elongated and fringed with minute setae, the endopodite is smaller.
- It captures the food and helps in feeding.

(7). SECOND MAXILLIPEDE:
- It is paired leaf like thoracic appendage situated beneath the first maxillipede one on either side.
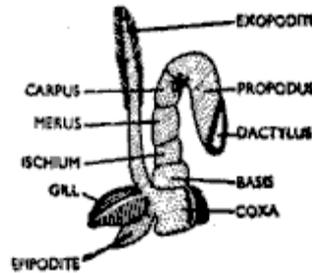
- The coxa bears second pair of primitive gills and a gill.
- The basis bears a slender exopodite which is provided with small setae.
- The endopodite is many jointed and made up of the following parts taken from the base upwards: ischium, merus, carpus, propodus and dactylus.
- It takes part in feeding and capturing the food.

(8). THIRD MAXILLIPEDE:

- These are paired thoracic appendages which resembles with the leg.
- The coxa is small and ring like, carries third pair of primitive gill.
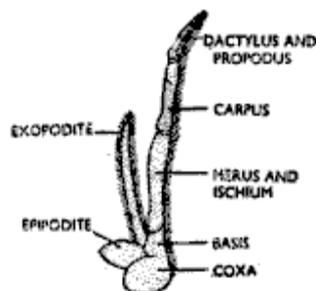
- The basis bears a long and slender exopodite and three segmented endopodite.
- In endopodite, the ischium and merus are fused, the carpus is free while the propodus and dactylus get compact.
- They help in capturing the food during feeding.

(9). WALKING LEGS:

- There are five pairs of walking legs which resembles very much in their structure except slight differences.
- Each leg is composed of seven segments, namely from the base as coxa, basis, ischium, merus, carpus, propodus and dactylus.
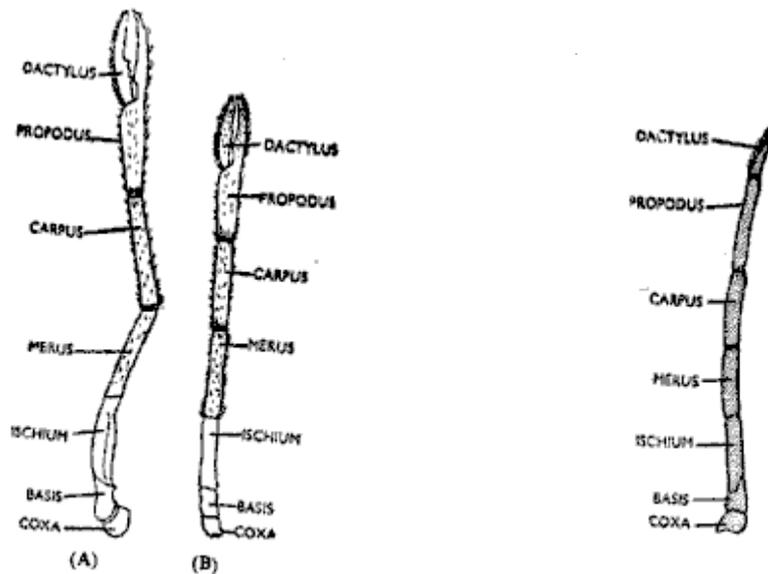
Chelate legs of Male and Female   Typical nonchelate legs

-       In first two pairs of legs the dactylus is formed with its inner serrated margin which are termed as chelate legs. They guide the food towards the mouth.
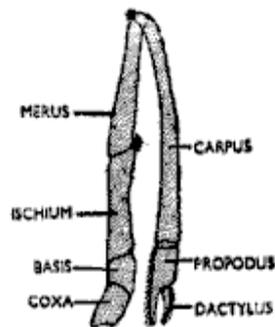
First Chelate leg

-       In male prawn, the second chelate leg is well developed and is more powerful than female.

-       In male the fifth pair of walking legs bears a male reproductive aperture on its coxa, where as in female the reproductive aperture is situated on the coax of third walking leg.

(10). PLEOPODS OR SWIMMERETS:

- There are six pairs of abdominal appendages, the first five of them are known as the **pleopods** or swimmerets and the sixth one is called the **uropod**.
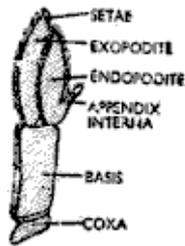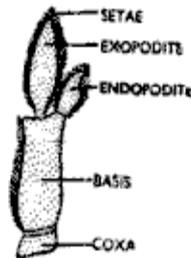
Typical appendage   **First appendage**   **Second appendage**
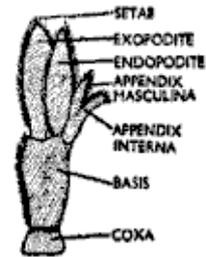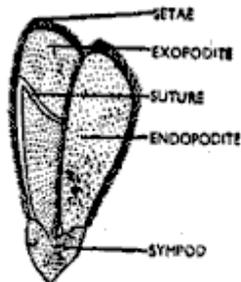
Uropod

5. Identification of food materials in gut of fish

Depending on their feeding adaptations and habits, different species of fish feed on different food organisms, such as phytoplankton, zooplankton, detritus, macrophytes, various macro invertebrates and fishes. Studies on the food and feeding habits of fishes are of very complicated nature and need much field and laboratory work. Direct observation on the feeding habits of a fish in its natural habitats is virtually impossible and thus to ascertain the exact nature of a fish food, the best way is to examine its gut contents.

Separation of stomach contents:

The region between the oesophagus and pyloric sphincter in the alimentary canal forms the stomach. Before removal of the stomach, the total length, sex and stage of sexual maturity of the fish should be recorded. The stomach is then carefully taken out, fixed in 5% formalin, dried between sheets of filter paper and slit open with a pair of scissors. If the stomach appears empty or contains only traces of food(less than 1.0 mg), it is rinsed with water directly into a Petri dish. If it contains a weighable quantity of food, the excess water is taken up using the absorbent tissue paper. The contents of the stomach are then weighed and washed in a Petri dish. Alternatively, the displacement volume of the stomach contents may be determined by dropping the lump into a known volume of 5% formalin in a graduated measuring jar. After recording the displacement volume of the stomach contents, latter is made into a known volume by adding more water. After stirring well, a substance of 1.0 ml is taken with a

graduated pipette and evenly spread over a counting slide. The contents are examined under a binocular or dissecting microscope. The food items identified and sorted into various taxonomic groups and the numerical percentage was estimated. Solitary fragments of crustaceans (appendages), polychetes (setae), mollusca(radula, mandible, shell parts) etc., are generally counted as full animals.

Various methods are used to analyse the gut content of which *Volumetric method* is often considered as more satisfactory and accurate measure for analysis of gut contents. In practice there are three ways of measuring the volume of food in the gut. They are:

(a). Eye estimation method: This is the easiest method used in the gut content analysis. Here, the contents of each sample are taken as unit and the various items are expressed as percentage volume by eye inspection. The contents of each gut are first vigorously shaken in a container so that they get mixed evenly. Next, a drop of the content is examined under microscope. The area occupied by each food item is estimated arbitrarily. At least ten such drops from each individuals should be examined and the average of each of the items be taken.

(b). Points (volumetric) method: This is basically similar to the eye estimation method. In this, instead of assessing by sight, each food item is allotted a certain number of points based on its volume. More over, the size of the fish and the fullness of the gut is also considered at the time of allotting the points.

(c). Displacement method: It is the most accurate volumetric method used in the gut content analysis. In this method, the volume of the each food item is measured by displacement of water in a graduated cylinder. This method is particularly suitable for carnivorous fishes. Where the quantity is small, the food items are distributed over a grid in the form of a layer of uniform and known thickness. The volume of each item is like other methods expressed as percentage of the total volume of the entire gut content.

6. Estimation of fecundity in fish

Fecundity has been considered as the number of ripening eggs in the female prior to spawning. It varies from species to species, depending on age, length, weight, environmental conditions, etc. Studies on fecundity are receiving much attention now a day as they play a key role in fish stock management. In many species of fish, the eggs are spawned over a short period so that the fish are mostly maturing or spent and those that are ripe and running are rare.

Method:

Collect the ovaries from a fish, weigh it and preserve them in a 5% formalin. After taking the weight, four sub-samples of known weights from different parts taken from the ovary and the different stages of ova are counted. The sub-samples were teased on the slide and a few drops of formalin were put to prevent the ova from getting dried up and the ova were counted under the microscope. The total number of ova in the entire ovary was calculated by the following formula.

$$F = \frac{S1 + S2 + S3 + S4}{SW1 + SW2 + SW3 + SW4} \times W$$

Where F is fecundity; S1, S2, S3, S4 are the number of ova in 4 sub-samples; SW1, SW2 , SW3, SW4 are the weights of 4 sub-samples and W is total weight of ovary in mg.

7. Estimation of length- weight relation in fish

The study of the length –weight relationship of fishes forms important aspects of fishery biology. One important aspect of the growth of a fish is the relationship between length and weight of its body. The statistical relationship between these two parameters helps in the identification of the analysis of the catches and breeding season. The analysis of these two parameters has usually been directed towards two different objectives, primarily towards describing mathematically the relationship between length and weight so that the expected weight for length of individual fish or the relevant group of individuals.

Method:

The fishes were studied this purpose should be grouped in to 10 mm class intervals. The length-weight relationship is calculated with the help of the formula:

$$W = ab^L$$

Where W is the body weight, L is total length and a & b are constants to be determined. a is a constant being initial growth, b is the grwoth co-efficient

The values of the constants a, b and regression coefficient (r) is calculated by the following formula:

$$b = \frac{\sum XY - n\,\bar{X}\bar{Y}}{\sum X^2 - n\,\bar{X}^2}$$

Where n = total no. of length groups

$\bar{x}$ = mean of x (length)

$\bar{y}$ = mean of y (weight)

The coefficient of co-relations r can be calculated

$$r = \frac{\sum XY - n\bar{X}\bar{Y}}{\sqrt{[(\sum X^2 - n\bar{X}^2)\,(\sum Y^2 - n\bar{Y}^2)]}}$$

If the value of r is found higher than 0.5, the length weight relationship is positively correlated and vice-versa

8. Collection of soil and water samples

(A). Sampling, Handling, Transport and Storage of soil samples:

The selection of sites and the method of collection of soil sample shall be depending entirely on the type of analysis to be performed and it is difficult to make generalization. However, composite sampling of soil is necessary to take representative sample from an area. The sediments from the water bodies are usually collected by employing a dredge. For this purpose Ekman dredge is commonly used and it suffices for surface layers.

Soil and sediment samplings can be collected in polythene bags and transported to the laboratory as early as possible. Some parameters should be determined immediately. For other parameters, the soil can be dried and stored. If immediately drying is not possible, the samples can be stored short periods at low temperature. The drying temperature for soil is critical, as many constituents change with change in temperature.

Soon after drying, stones and other similar objects are picked up and the soil is ground in a mortar to break up aggregates or lumps, taking care not to break actual soil particles. The soil is then passed through a sieve. For sieving purpose stainless steel or nylon sieves are preferable.

(B). Sampling, Collection, Handling and Preservation of water samples:

The sampling location of the water body shall depend upon the character of the water body. In a lake or a wide river many

sampling sites should be selected at various corners. If the lake is stratified, three vertical samples at one site (surface, middle and bottom) shall be required. In shallow ponds, only surface and bottom samples or a sample from 0.5 to 0.7 m depth shall also suffice. In a stream which is narrow and rapidly moving the water shall be thoroughly mixed laterally and vertically hence only one sampling point need to be selected at each location along the stream.

The water samples may be collected by using a water sampler or by using glass or polyethylene bottles. For collection of a sample from the bottom, where water is shallow (around one meter) the sample can be collected by lowering a closed bottle to the bottom. Opening and closing it thereby hand and bringing at the surface. But in this case, the surface samples must be collected first to avoid the disturbance caused by loose sedimentation. Prior to the collection, the sample bottle must be rinsed thoroughly with the sample water even if it is precleaned.

Accurate sampling should be followed by correct handling and preservation of the sample to obtain the reliable results. Some of the important points one should be taken care are:

1. Immediately after collection, clearly label each sample bottle with water proof ink and record the relevant details for each sample.
2. Temperature of water should be immediately recorded.
3. The samples for chemical analyses should preferably be collected separately.

4. The sample should be taken to the laboratory as early as possible; it should be protected from direct sunlight during transportation.

Since most of the parameters change with time, it is imperative to preserve them in a suitable preservative prior to analyses. Dissolved oxygen, Free CO2, Alkalinity and pH quickly change with the time, their estimation is to be carried out in the field only.

9. PHYSICO-CHEMICAL PARAMETERS OF WATER:

In fisheries point of view, Water is the primary requisite to support the aquatic life. Maintenance of a healthy aquatic environment is the secret of success in pond culture activities. The physical, chemical and biological factors play an important role in governing the production of fish food organisms and fish production in the pond. Water not only plays an important role in the production of fish, but also it helps in the survival and growth of the fish. Hence, fish farmers should take necessary care to maintain the optimal conditions in the culture ponds. Some of the important physical and chemical parameters , that are to be monitored are estimated as per the procedures mentioned below:

(i). pH (Potentia Hydrogeni):

pH measures the hydrogen in concentration in the water. It is measured on a log scale and equals to negative log10 of hydrogen ion concentration. A neutral solution has a pH of 7 while a pH less than 7 renders it acidic; and pH more than 7, alkaline. Water is slightly alkaline in condition, with the optimal range of 6.5 to 8. Less than 5 and more than 10 pH is lethal to fish. Generally, the pH of the pond water always undergoes a diurnal change. It is alkaline during the day time and slightly acidic just before day break. The difference in pH from morning to evening should not be more than 0.5. When pH increases, ammonia and nitrites become toxic and decrease the H2S becomes more toxic.pH below 6.5 and above 8.5 is responsible for reduction of growth.

The pH can be obtained by using either colorimetric method employing various indicators or by using sensitive electrometric methods employing hydrogen ion sensitive electrodes in pH meters. However, the latter gives an accurate and quick measure of the pH.

THE pH METER

There are various models and kinds of pH meters manufactured by a number of companies. Essential feature of all the pH meters is to use a hydrogen sensitive electrode, called indicator electrode, and a colomel reference electrode. The indicator electrode is generally made up of a highly sensitive and thin glass membrane. Some pH meters combine the both these electrode in a single electrodes, while in the others they remain separate Most pH meters also possess a temperature compensation system to avoid the differences arising due to the different temperatures.

PREPARATION OF BUFFERS

1. Potassium hydrogenyphthalate buffer Dissolve 10.2g of potassium hydrogenyphthalate in water to prepare 1000ml of buffer. The pH value of this buffer at 20^0C is 4.

2. Phosphate buffer:
    Dissolve 3.40g of KH_2PO_4 and 4.45g of $Na_3HPO_4\ 2H_2O$ in water to prepare 1000ml of buffer. The pH of this buffer at 20^0Cis 6.9.

3.   Borax bufer:
     Dissolve 3.8g of $Na_4B_7O_{10}H_2O$ in water to prepare 1000ml of bufur. The pH value of this bufffer at 20^0C is 9.22

PROCEDURE:

As the pH meters are made in different models and by different companies. It is advisable to follow the instructions supplied by the manufacturer. However, essential aspect to use all the pH meters is to calibrate it with suitable buffers. Ready buffers can be purchased from the market. Set the pH meter with the buffer whose value is near to the expected pH of the sample. pH value now can be obtained directly by immersing the electrode in sample.

(ii) Temperature:

Temperature is measure of the hotness of any material. This measurement of temperature in water is important basically for its effects on the chemistry and biological reaction in the organisms. It is also important in the determination of pH, conductivity and saturation level of gases in water. It affects the fish migration, reproduction and distribution. It depends upon climate, sun light and depth of the pond. Fish possess a well defined limits of temperature tolerance with the optimal being $20 – 32^0C$. Wide fluctuations in temperatures affect the survival of the fish. At low temperatures the food consumption of fish decreases and gasses are produced at high temperatures.

PROCEDURE:

Surface water:

The surface water temperatures are best determined by the use of mercury thermometers upto the desired accuracy. In most ecological situations depending upon the study the accuracy may vary from 0.1 to 1.0^0C. To minimize the errors, it is essential to calibrate thermometer with another thermometer of known accuracy. While taking the reading, our eyes should be at right angles to the mercury thread. The scale of the thermometer should be immersed in the water upto the level of mercury in the capillary column.

(iii). CONDUCTIVITY:

Conductivity denotes the capacity of a substance or solution to conduct the electric current. Conductivity if measured of a cube with each side of 1 cm at 25^0C is called the specific conductance. In water, it is the property caused by the presence of ions. It is generally measured with the help of a conductivity meter.

CONDUCTIVITY METER:

The main feature of the most conductivity meters is a conductivity cell containing electrodes of platinum coated with Pt black or carbon. These electrodes are mounted rigidly and placed parallely at a fixed distance. Most conductivity meters work on the principle of wheatstone bridge in which the cell forms an arm of the bridge.

PROCEDURE

For use of the conductivity meter, follow the instruction supplied by the manufacturer.

The conductivity depends upon the area of the metallic plates in the cell and the distance between them. To convert, the observed conductance in specific conductance the values are multiplied by a factor called cell constant which is generally supplied by the manufacturer. After taking the reading, note down the temperature of the sample and fin out the factor from Table 1 to convert the value at 25^0C.

Table 1 Factors for converting the values of conductivity at 25^0C (After golterman et.al.1978)

^{0}C	Factor	^{0}C	Factor	^{0}C	Factor
32	0.89	22	1.06	12	1.30
31	0.90	21	1.08	11	1.33
30	0.92	20	1.10	10	1.36
29	0.93	19	1.12	9	1.39
28	0.95	18	1.14	8	1.42
27	0.97	17	1.16	7	1.46
26	0.98	16	1.19	6	1.50
24	1.00	15	1.21	5	1.54
25	1.02	14	1.24	4	1.58
23	1.04	13	1.27	3	1.62

Calculation

Conductivity = observed conductance x cell constant x temperature factor at 25^0 C

(iv). Dissolved Oxygen:

The presence of dissolved oxygen is essential to maintain the higher forms of biological life and to keep the proper balance of various populations thus making the water body healthy. The chemical and biochemical processes undergoing in a water body are largely dependent upon the presence of Oxygen. The pond water gets oxygen mainly through interaction of atmospheric air on the surface water of the pond and by photosynthesis. It is produced only during the day time, reaches a maximum at 3 PM, and then gradually decreases upto early morning. During over cast days, the production of dissolved oxygen during the day is less and during the subsequent nights it decreases drastically. The optimal dissolved oxygen is 5 – 8 ppm. If less than 5 ppm the growth rate decreases and the fishes are prone to get diseases. If it is less than 1 ppm, results into death. More than 15 ppm results into a gas bubble disease in fish.

REAGENTS

A. Sodium thisulphate, 0.025 N

Dissolve 24.82 g of Na_2 of $Na_2 S_2 O_3$. $5H_2 O$ in a preboiled water and make up the volume to 1 liter. Add a pellet of NaOH or 0.4 g of borax as a stabilizer. This is 0.1 N stock solutions. Dilute it to 4 times to prepare 0.025 N solution (250 → 1000 ml). Keep in a brown glass bottle.

B. Alkaline potassium iodide solution

Dissolve 100g of KOH and 50 g of KI in 200ml of preboiled distilled water.

C. Manganous sulphate solution

Dissolve 100g of $MnSO_4$ 4 H_2 O in 200 ml of distilled water and heat to dissolve maximum salt; filter after cooling.

D. Starch solution.

Dissolve 1g of starch in 100 ml of distilled water, warm for complete dissolution.

E. Sulphuric acid, conc.(Sp.gr.1.84)

PROCEDURE:

Fill the sample in a glass stopper bottle of known size (100-300ml). Care should be take to avoid any kind of bubbling and trapping of the air bubbles after placing the stopper. Add 1 ml of each $MnSO_4$ and alkaline KI solutions well below the surface through the walls. Use always separate pipettes for these two reagents. A brown precipitate will appear indicating the presence of Oxygen. Shake the contents well by repeatedly inverting the bottle and keep for sometime settling down the precipitate in case the precipitate is not settling properly, again shake the contents and keep. If the titration is to be prolonged for few days, the sample can be stored at this stage with the precipitate. Add 2 ml of concentrated H_2 SO_4 and shake well to dissolved the precipitate. Remove either the whole contents or part of them (50-100 ml) in a conical flask of titration, avoiding

any further mixing of oxygen through bubbling. Titrate the contents, within one hour of dissolution of the precipitate with sodium thiosulphate solution using a few drops of starch as an indicator. At the end point, the initial dark blue-black colour changes to colourless.

CALCULATION

When the whole contents have been titrated:

$$\text{Diss. Oxygen, mg/L} = \frac{(mlXN) \text{ of sodium thisoulphate } x8x1000}{V_1 - V}$$

When only a part of the contents has been titrted:

$$\text{Diss. Oxygen, mg/L} = \frac{(mlXN) \text{ of sodium thiiosulphate } x\ 8x\ 1000}{V_2 \dfrac{(V_1 - V)}{V_1}}$$

where V_1 = Volume of sample bottle
V_2 = Volume of content titrated
V = Volume of $MnSO_4$ and KI added (2ml)

Sl.No	Volume of the sample	Burette reading		Avaerage
		Intial	Final	
1				
2				

(V). Free Carbon Dixoide:

Free carbondioxide in the waters accumulates due to microbial activity and respiration of organisms. This imparts are acidity to the waters because of the formation of carbonic acid. Generally it is produced during the respiration and consumed during the photosynthesis. CO_2 is less during the day time and more at night. The optimum level of CO_2 is 5 ppm. At high CO_2 levels., pH decreases, CO_2 accumulated in the blood of the fish and water becomes more acidic. The fish becomes sluggish, loss of resistance occur. Free CO_2 is determined by titrating the sample using a strong alkali of pH 8.3.

REAGENTS

A Sodium hydroxide, 0.05 N

Dissolve 40 g of NaOH in boiled CO_2 free distilled water and make up the volume to 1 litre. Filter the solution through a sintered glass filter to remove any Na_2CO_3. This is 1.0 NaOH solution. Store it in a ploythene air tight bottle. Dilute this solution to 20 times to prepare 0.05 N solution only at a time of titration. Standardize the diluted solution with H_2SO_4. HCL or oxalic acid.

B Phenolphtalein indicator

Dissolve 0.5 g of phenolphtanlein in 50 ml of 95% ethanol and add 50 ml of distilled water. Add 0.05 N CO_2 free NaOH solution dropwise, until the solution just turns faintly pink.

PROCEDURE

100ml of the sample in a conical flask and add a few drops of phenolphtalein indicator. The colour change to pink indicates the absence of free CO_2. In case the sample colourless, titrate it with 0.06 N NaOH. At the end point a pink colour will appear.

CALCULATION

$$\text{Free } CO_2, \text{mg/L} = \frac{(\text{mlXN}) \text{ of NaOHX100X44})}{\text{ml sample}}$$

Sl.No	Volume of the sample	Burette reading		Avaerage
		Intial	Final	
1				
2				

(vi). Total alkalinity, carbonates and bicarbonates:

Alkalinity of the water is its capacity to neutralize a strong acid and is characterized by the presence of all hydroxyl ions capable of combining with the hydrogen ion. Alkalinity in natural waters is due to free hydroxyl ions and hydrolysis of slats formed by weak acids and strong bases such carbonates and bicarbonates. In fish ponds, the optimal level of total alkalinity is 40 – 150 ppm.

REAGENTS

A. Hydrochloric acid 0.1 N

Dilute 12 N concentrated HCL (sp.gr. 1./8) to 12 times (8.34 in 100 ml) to get 1 0 N HCL. Further dilute it ot 10 time prepare 0.1 N HCL (100 in 1000ml) Standardize it against sodium carbonate solution.

B. Sodium, Carbonate , 0.1 N

Dissolve 5.300 g of Na_2CO_3 (predried at 250^0C for four hours in distilled water to prepare 1 liter of solution.

C. Methyl orange indicator, 0.5%

Dissolve 0.5 g of methyl orange indicator in 100 ml of distilled water.

PROCEDURE:

Take 100ml of sample in conical flask and add drops of phenolphthalein in it. If the solution remains colourless, the phenolphthalein alkalinity (PA) is zero (indicating absence of carbonates) and total alkalinity with methly orange is only determined., if the colour changes to pink after addition of phenolphthalein, titrate it with 0.1 N HCL unit the colour disappears at end point. This is phenolphthalein alkalinity (PA). Now add 2-3 drops of methyl orange to the same sample and continue the titration further, until the yellow colour changes to pink at the end point. This is total alkalinity (TA).

Sl.No	Volume of the sample	Burette reading		Avaerage
		Intial	Final	
1				
2				

Calculations:

PA as CaCo$_3$ mg/1= $\dfrac{(AXN) \text{ of } HCLX1000X50}{ml/sample}$

TA as CaCo3 mg/L= $\dfrac{(BXN) \text{ of } HCL \ X1000X50}{ml \ sample}$

where A=ml of HCL used only with phenolphthalein
B= ml of HCL used with phenolphthalein and methyl orange
i.e.total HCL used with both the indicators.

(vii). Phosphates:

The phosphate is generally considered as the critical nutrient for the growth of algae in water. The enrichment of this nutrient leads to the process of eutrophication. The most important sources of phosphates are the discharge of domestic sewage, detergents and agricultural run-off.

REAGENTS:

A. Ammonium molybdate solution

a. Dissolve 25.0 g of ammonium, molybdate in 175 ml of distilled water.

b. Add 280 ml of concentrated H_2SO_4 to 400ml of distilled water and cool

Mix the two solutions (a) and (b) and make up of the volume to 1 litre.

B. Stannous cholride solution

Dissolve 2.5 g of stannous chloride in 100 ml glycerol by heating on a water bath.

C. Standard Phosphate solution

Dissolve 4.388 of predried anhydrous potassium hydrogen phosphate/ Sod, Phosphate K_2HOP_4 in distilled water and make up the volume to 1 litre. Dilute this solution to 100 times (10-1000ml). This is standard phosphate solution containing 10 mg P/L (1=0.01 mg P)

PROCEDURE

Take 50 ml of filtered, clear and colourless sample in a conical flask. If the sample is having colour and colloidal impurities, remove them by adding a spoonful of activated charcoal and filtering. Now add 2 ml of ammonium, molybdate

solution and 5 drops of stannous chloride reagent. A blue colour will appear in presence of phosphate. Take the optical density reading at 690 mm on spectrophotometer using a distilled water blank with the same amount of the chemicals. Readings on the spectrophotometer should be taken after 5 minutes but before 12 minutes of the addition of the last reagent. Find out the concentration of phosphate with the help of the standard curve.

Prepare the standard curve in the range of 0.0 to 1.0mg/L of PO_4-P at the interval of 0.1, following the same method described for the sample.

Prepration of various dilutions of phosphorus for the standard curve

ml of standard Solution	dilute to (ml)	concentrationof $PO4$ -P, mg/1
5.0	50	1.0
4.5	50	0.9
4.0	50	0.8
3.5	50	0.7
3.0	50	0.6
2.5	50	0.5
2.0	50	0.4
1.5	50	0.3
1.0	50	0.2
0.5	50	0.1

(viii). Nitrates :

Like phospharus, it is also one of the critical nutrients for the growth of algae and helps accelerating the eutrophication. Domestic sewage, natural run-off and agricultural wastes are the important sources of it. The determination of nitrate in drinking water is of prime importance of the disease methomoglobinemia caused by its excessive presence.

REAGENTS:

A. Phenol disulphonic acid

Dissolve 25.0 g of white pure phenol in 150ml of cone. H_2SO_4 and add 75ml of fuming H_2SO_4. Heat for about 2 hours on a steam bath and keep in a dark bottle. In place of 75ml fuming sulphuric acid, 85ml cone. H_2SO_4 can also be added.

B. Silver sulphate solution.

Dissolve 4.4 g of Ag_2SO_4 in distilled water and make up the volume to 1 litre.

C. Liquid ammonia, 30%,

D. Standard nitrate solution (mg N,L)
Dissolve 0.722 g of KNO_3 in distilled water to prepare 1 litre of solution. This solution contains 100 mg N/L. Dilute to 100 times of prepare a solution having 1 mg N/L (10-100 ml).

PROCEDURE

Take 50ml of filtered or an aliquot containing not more than I mg/L of NO_3-N in a conical flask. To remove the interference of chloride, add an equivalent amount of sliver sulphate solution (1 mg/L of CL- = 1 ml Ag SO_4 solution. Heat slightly and filter the precipitate of Ag Cl. Evaporate the filtrate in a porcelain basin to dryness, cool and dissolve the residue in 2ml phenol di- sulphonic acid. Dilute the contents to 50 ml and add 6 ml of liquid ammonia to develop a yellow colour. Take the absorbance of this colour at 410 nm and calculate the NO_4- N from the standard curve.

Prepare standard curve between concentration and absorbance from 0.0 to 1.0 mg/L of NO_3-N at the interval of 0.1. Find out the absorbance of the standards using the same procedure described for the sample above except the removal of chlorides by addition of sliver sulphate.

Sl. No	Optical Density Standard	Optical Density Experimental

(ix). Hardness:

Hardness is the property of water which prevents the lather formation with soap and increase the boiling point of waters. The major cations imparting hardness are calcium and magnesium. The anions responsible for hardness are bicarbonate. Carbonate, sulphate and chlorides. Hardness is temporary if it is associated mainly with carbonates and bicarbonates; and permanent, if with sulphates and chlorides. Water with less than 40 ppm is soft and more than 40 ppm is hard water. The pond water with a hardness of 15 ppm or more is satisfactory for growth of fishes.

REAGENTS

A. EDTA solution, 0.01M
Dissolve 3.723 g of disodium salt of EDTA in distilled water to prepare 1 litre of solution. Store in polyethylene or pyrex bottle.

B. Buffer solution
a. Dissolve 16.9 of disodium chloride (NH4 cl) in 1.43 ml of concentrated ammonium hyroxide (NH_4OH).
b. Dissolve 1.179 g of disodium EDTA and 0.780 g of $MgSO_4$. $7H_2O$ in 50ml distilled water.
c. Eriochrome Black T indicator
Grind 0.40 g of Eriochrome Black T with 100 g NaCL (A.R.)

C. Sodium sulphide solution – Dissolve 5.0 g of $Na_2S.9H_2O$ or 3.7 g $Na_2S.5H_2O$ in 100 ml of distilled water and store in a tightly closed bottle.

Procedure:

Take 50 ml of sample in a conical flask , In case of sample with high hardness a small aliquot may also be taken. Add 1 ml of buffer to this. If the sample is having higher amounts of heavy metals, add 1 ml of Na_2S solution. Add approximately 100 mg of Eriochrome Black T indicator, the solution will turn wine- red. Titrate the contents with EDTA solution; the colour change to blue at the end point.

Sl.No	Volume of the sample	Burette reading		Avaerage
		Intial	Final	
1				
2				

Calculation:

Hardness, as $CaCO_3$ mg/L $= \dfrac{\text{ml of EDTA used X1000}}{\text{mL of sample}}$

(ix- a). Calcium:

Calcium is one of the most abundant elements found in the natural water. It is an important ion in imparting the hardness to the waters. At high pH much of its quantities may get precipitated as $CaCO_3$

REAGENTS:

A. EDTA solution , 0.01 M
    See determination of hardness

B. Sodium hydroxide, 1 N
    Dissolve 40g of NaOH in distilled water to prepare 1 litre of solution.

C. Murexide indicator.
    Grind 0.2g of ammonium purpurate with 100g of NaCl (A.R.)

PROCEDURE

Take 50ml of sample in a conical flask. In case of the sample with higher alkalinity, a smaller aliquot of sample should be taken Add 2.0ml of NaOH solutin and approximately 100 mg of murexide in the sample; a pink colour will develop. Titrate the content with EDTA solution until the pink colour changes to dark purple. As there is no sharp end point, for better judgement compare the end point colour with that of the purple colour obtained after distilled water blank titration end point.

Sl.No	Volume of the sample	Burette reading		Avaerage
		Intial	Final	
1				
2				

Calculation:

$$\text{Calcium mg/L} = \frac{(x) \text{ ml EDTA use} \times 400.8}{\text{ml sample}}$$

(ix -b). Magnesium:

Magnesium also occurs in all kinds of natural waters, but its concentration remains generally lower than the calcium. Like calcium, it is also one of the important cations imparting hardness to the waters.

REAGENTS:

A. EDTA 0.01M
B. Buffer solution
C. Eriochrome Black T indicator.
D. Sodium hydroxide , 1 N
E. Mure ide indicator.

PROCEDURE

Magnesium is determined as the difference between the Ca+Mg titration and the titration alone for Ca. Perform the titration for Ca as given previously and find out the volume of EDTA used. Also find out the volume of EDTA A used for Ca+Mg titration following the method given for hardness using the same of volume of sample as used in Ca determination alone.

Sl.No	Volume of the sample	Burette reading		Avaerage
		Intial	Final	
1				
2				

CALCULATION

$$\text{Magnesium, mg/L} = \frac{(y-x) \times 400.8}{\text{ml sample} \times 1.645}$$

where x = EDTA used for Ca determination

y = EDTA used for hardness (Ca + Mg)

determination using the same volume of sample as used for Ca.

(x) .Chlorides:

Chloride occurs naturally in all types of waters. In natural fresh waters, however, its concentration remains quite low. The most important source of chloride in natural waters is the discharge of sewage. In very high concentration it gives a salty taste to the water.

Reagents:

A. 0.02 N Silver nitrate solution – Dissolve 3.400 g of pre dried Ag NO_3 (A.R) in distilled water to prepare a 1 litre of solution. Keeping in a dark glass bottle.

B. 5% Potassium chromate indicator – Dissolve 5 g of K2Cr O4 solution in 100 ml of distilled water.

Procedure: Take 50 ml of water sample in a conical flask and add 2 ml of K_2CrO_4 solution. Titrate the contents against 0.02 N $AgNO_3$ until a persistent reddish brown tinge appears.

Sl.No	Volume of the sample	Burette reading		Avaerage
		Intial	Final	
1				
2				

Calculation

$$\text{Chlorides in mg/L} = \frac{(ml \times N) \text{ of } AgNO_3 \times 1000 \times 35.5}{ml \text{ of sample}}$$

10. Study of Soil types and Estimation of Soil parameters

The soil is developed by the weathering of the rocks present in nature and differentiated into horizons of various heights and characters. The soil is always different from its parent material as the morphological, chemical and biological characters are concerned.

Soils are very productive contains adequate amounts of all essential elements in the form of readily available to the biota. For good aquaculture practices, the soil should always be in good physical and chemical conditions. The soil productive capacity in most cases, can be evaluated satisfactorily by determining physical , chemical and microbial properties of the soil.

(i). pH

pH of soil is the measure of 'hydrogen ion activity' and depends largely on the relative amounts of the adsorbed hydrogen and metallic ions. It is a good measure of the intensity of acidity and alkalinity of a soil-water suspension, and provides a good identification of the soil chemical nature.

pH soil suspension highly depends on the soil water ration. Determination of pH at moisture saturation level and 1:5 soil suspension is most common.

Apparatus and Reagents

THE pH METER

There are various models and kinds of pH meters manufactured by a number of companies. Essential feature of all the pH meters is to use a hydrogen sensitive electrode, called indicator electrode, and a colomel reference electrode. The indicator electrode is generally made up of a highly sensitive and thin glass membrane. Some pH meters combine the both these electrode in a single electrodes, while in the others they remain separate Most pH meters also possess a temperature compensation system to avoid the differences arising due to the different temperatures.

PREPARATION OF BUFFERS

4. Potassium hydrogenyphthalate buffer Dissolve 10.2g of potassium hydrogenyphthalate in water to prepare 1000ml of buffer. The pH value of this buffer at 20^0C is 4.

5. Phosphate buffer:
Dissolve 3.40g of KH_2PO_4 and 4.45g of $Na_3HPO_4 2H_2O$ in water to prepare 1000ml of buffer. The pH of this buffer at 20^0C is 6.9.

6. Borax buffer:
Dissolve 3.8g of $Na_4B_2O7 10H_2O$ in water to prepare 1000ml of buffur. The pH value of this bufffer at 20^0C is 9.22

Procedure

To determine the pH at the moisture saturation level. Take about 50g of soil in a beaker. Add small portions of distilled water, without stirring the soil, until a glistering appears at the surface. Now make a uniform paste of soil by stirring with the help of a glass rod.

For determination of pH of soil in 1:5 soil suspension, take 20g of soil and add 100ml of distilled water stir for about an hour at regular intervals.

As the pH meters are made in different models and by different companies. It is advisable to follow the instructions supplied by the manufacturer. However, essential aspect to use all the pH meters is to calibrate it with suitable buffers. Ready buffers can be purchased from the market. Set the pH meter with the buffer whose value is near to the expected pH of the sample. pH value now can be obtained directly by immersing the electrode in sample.

Expression of Results

Results are expressed directly in pH units associated with the specific dilution of soil suspension, e.g. pH in 1:5 soil suspension.

(ii). Alkalinity:

Like water, total alkalinity is determined by the direct

titration of the soil solution with a strong acid using methyl orange as an indicator.

REAGENTS

A. Hydrochloric acid, 0.1 N

Dilute 12 N concentrated HCL (sp.gr. 1./8) to 12 times (8.34in 100 ml) to get 1 0 N HCL. Further dilute it ot 10 time prepare 0.1 N HCL (100 in 1000ml) Standardize it against sodium carbonate solution.

B. Sodium carbonate, 0.1N

Dissolve 5.300 g of $Na_2 CO_3$ (predried at 250^0C for four hours in distilled water to prepare 1 liter of solution.

C. Methyl orange indicator, 0.5%

Dissolve 0.5 g of methyl orange indicator in 100 ml of distilled water

Procedure:

Prepare 1 : 5 soil suspension by shaking 20 g of soil in 100 ml of aerated distilled water for about one hour. Filter the suspension through Whatman No. 50 filter paper using Buchner funnel and vaccum pump. Take 100 ml of this soil solution. Add 2-3 drops of methyl orange and titrate with 0.1 NHCl. At the end point the colour will change to pink.

Calculations:

PA as CaCo$_3$ mg/1 = $\dfrac{(AXN) \text{ of } HCLX1000X50}{ml/sample}$

TA as CaCo3 mg/L = $\dfrac{(BXN) \text{ of } HCL\ X1000X50}{ml\ sample}$

where A = ml of HCL used only with phenolphthalein
B = ml of HCL used with phenolphthalein and methyl orange
i.e.total HCL used with both the indicators.

CONTENTS

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