ZOOLOGY VIRTUAL LAB 2
(INVERTREBRATE MOUNTINGS PART II)

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PRACTICAL 1:

MOUNTING OF MOUTH PARTS OF HONEY BEE

AIM: To mount and study the mouth parts of Honey bee.

BACKGROUND INFORMATION:

Mouth parts are the appendages grouped around the mouth. The 'primitive' arrangement of mouthparts is seen in the Honey bee - here they are chewing and lapping type. They are modified for collecting nectar and pollen and work in the vertical plane.

REQUIREMENTS:

Honey bee, 10 % KOH, Scalpel, Watch glass, Test tube, Holder, Burner, Slides, Filter paper, Dissecting microscope, etc.

PROCEDURE:

1. Cut the head part of honey bee with the help of Scalpel.
2. Put the removed head in the test tube and add 10% KOH.
3. Attach the holder to the test tube and boil for 2-3 mins on flame.
4. Put the test tube content in the watch glass.
5. Keep the treated head on slide and put another slide on it and gently press so the inner parts will come out.
6. Observe it under dissecting microscope.

OBSERVATIONS AND RESULT:

Mouthparts consist of labium epipharynx, mandibles and maxillae.

The labium lies below the clypeus. The mandibles are smooth and are situated on either side of the labium. They are used for molding the wax and making the honeycomb. The labium has submentum, mentum, paraglossa with a long labial palp. The glossa has labellum (honey spoon) as its tip. The maxillae fit over the mentum and bear maxillary palps. They are sensory in function. The maxillae and labial palps forma tube enclosing the glossa, which moves up and down while collecting the nectar.

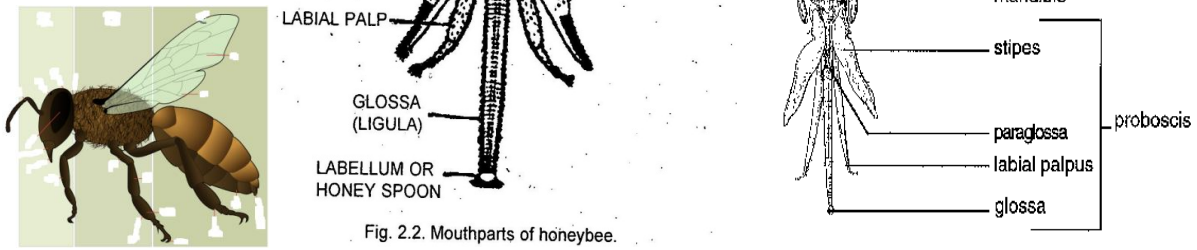


Fig. 2.2. Mouthparts of honeybee.

PRACTICAL 2:

MOUNTING OF STING APPARATUS OF HONEY BEE

AIM: To mount and study sting apparatus of Honey bee.

BACKGROUND INFORMATION:

A **stinger** or string apparatus is a sharp organ found in various animals (typically arthropods) capable of injecting venom, usually by piercing the epidermis of another animal. An insect bite or sting is a break in the skin or puncture caused by an insect and complicated by introduction of the insect's saliva, venom, or excretory products. Specific components of these substances are believed to give rise to an allergic reaction. Stinging insects produce a painful swelling of the skin, the severity of the lesion varying according to the location of the sting and the identity of the insect. Many species of bees and wasps have two poison glands, one gland secreting a toxin in which formic acid is one recognized constituent, and the other secreting an alkaline neurotoxin; acting independently, each toxin is rather mild, but when they are injected together through the sting, the combination has strong irritating properties. Bee dies after stinging because the stinger remains in the skin, attached to some of the bee's vital organs.

REQUIREMENTS:

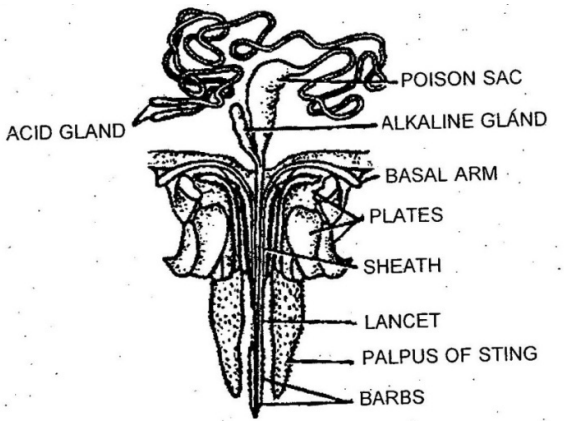
Honey bee, Needle, Forcepss, Slide, Cover slip, Water, Glycerine, Dissecting microscope, etc.

PROCEDURE:

1. Cut the posterior part of Honey bee with Scissor.
2. Put it on slide and with the help of needle and Forcepss remove the turgal and sternal plates.
3. Loose the tissue and locate brownish part.
4. Discard the other debris and place a drop of glycerine.
5. Put the cover slip and observe it under low power objective.

OBSERVATIONS AND RESULT:

The sting apparatus is modified ovipositor in worker bees. It consists of three parts viz., gonapophysis, stylets and poison canal. There are two stylets which are articulated along their length. The stylets are laterally covered by plates which are named as triangular plate, quadrate plate and oblong plate. At the tip of the stylet a pointed lancet is present which is provided with barbs. Attached to the stylet at the posterior region is a poison sac into which the alkaline gland opens. The three plates act like a spring and help in inserting the sting.



PRACTICAL 3:

MOUNTING OF LEGS OF HONEY BEE

AIM: To mount and study legs of Honey bee.

BACKGROUND INFORMATION:

In Honeybee, there are three pairs of legs performing different functions. They are Prothoracic, Mesothoracic and Metathoracic legs. Each leg is made up of five parts namely coxa, trochanter, femur, tibia and tarsus which terminate with a pair of claws. Each pair of legs differs in size and shape from the other two pairs and is jointed into six segments, with a pair of claws at the tip which help the insect to cling to surfaces. The leg can be flexed at any of the six joints. Its primary function is to help the bee to walk and run, but various parts also serve special purposes other than locomotion. For example, the brushes on the inner surface of the fifth segment, (the tarsus) of the two front legs are used for sweeping pollen and other particles from the head, eyes and mouth parts. The same tarsi of the mid-legs serve as brushes for cleaning the thorax, while the spines found at the end of the fourth sections (tibiae) are used for removing the pellets of pollen and for cleaning the wings. Two important parts to note on the legs are the antenna cleaners on the front legs and the pollen baskets on the hind legs.

REQUIREMENTS:

Honeybee, Forcepss, Slide, Water, Glycerine, Dissecting microscope, etc.

PROCEDURE:

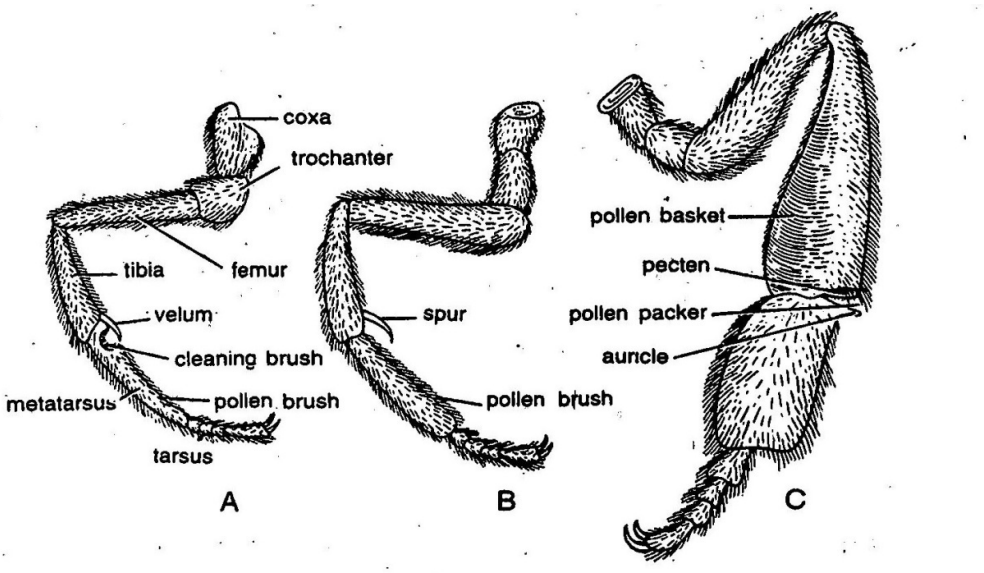
1. Locate first pair of appendages in thoracic region and with the help of Forcepss remove it from its base.
2. Place the leg on slide and observe it under dissecting microscope.
3. Follow same procedure for 2^{nd} and 3^{rd} pair of appendages.
4. Compare the structures of all three legs.

OBSERVATIONS AND RESULT:

Prothoracic leg - It has clusters of bristles in a row on the tibia, forming an eye brush. At the distal end of the tibia a movable spine is present called velum which closes over a notch on the tarsus and it is called antenna comb. Long bristles o the tarsus form the pollen brush which is used for the removal of pollen.

Mesothoracic leg - It is also has a pollen brush on the tarsus and at the end of the tibia it has a spur or a spine, which is used for removing pollen from the pollen basket and wax from the abdomen.

Metathoracic leg - It has a large tibia which contains the pollen basket whose cavity is filled with bristles to contain the pollen. At the distal end the tibia has thick bristles called pectin below which a flat plate called auricle is present. Pectin and auricle constitute the pollen packer. It is also used for removing wax from the abdomen. The outer surface of tarsus has pollen brush and the inner surface of tarsus has pollen comb. The pollen comb removes the pollen from the body and fills it in the pollen basket.



.86. Legs of honeybee. A – Prothoracic leg; B – Mesothoracic leg, C – Metathoracic leg.

PRACTICAL 4:

MOUNTING OF SEPTAL NEPHRIDIA OF EARTHWORM

AIM: To mount and study of Septal Nephridia of Earthworm.

BACKGROUND INFORMATION:

The nephridium (plural *nephridia*) is an invertebrate organ which occurs in pairs and performs a function similar to the vertebrate kidney. Nephridia remove metabolic wastes from an animal's body. They are present in many different invertebrate lines. There are two basic types: metanephridia and protonephridia, but there are other types. Metanephridium are coiled excretory tubules in most annelids.

In earthworm structurally there may be three types of metanephridia, the septal, the pharyngeal and the integumentary. The septal nephridia will be described as their structure is typical. They are present on both the anterior and the posterior surfaces of each septum from the septum between 15/16 segments onward.

REQUIREMENTS:

Earthworm, tray, water, glycerine, eosin, Forcepsss, needle, filter paper, slide, cover slip, compound microscope, etc.

PROCEDURE:

1. Mark the dorsal side by locating the mid dorsal black line and pin the earthworm in wax tray containing water.
2. Cut open the animal and observe very small coiled thread like structures attached to all the septa behind the 15^{th} segment.
3. Cut a part of any septum behind the clitellum and collect in onto a slide with the help of BB Forcepsss.
4. Observe the slide under a dissecting microscope and you will find a number of coiled nephridia. Separate them with Forcepsss and try to keep only one complete nephridium.
5. Stain it with eosin by pouring a few drops of stain on the nephridium.
6. Pour a few drops of water on the slide to wash the extra stain. Place a cover slip. Do not press it.
7. Observe the slide under a compound microscope at low power.

OBSERVATIONS AND RESULT:

These nephridia remain attached to all the septa behind the 15^{th} segment. They are known as the septal nephridia. This coiled, tubular structure made up of four parts: funnel, neck, body and terminal duct.

Funnel: This is the inner end of the Nephridium which is funnel shaped. It has a ciliated margin. It opens into the body cavity through an opening called as Nephrostome.

Neck: This is a short, narrow, coilated tube which connects the funnel with the body of the nephridium.

Body: This is the largest part of the Nephridium. It consists of two parts, a short straight lobe and a long, spirally twisted loop.

Terminal Duct: The last part of the Nephridium. It is narrow tubular structure.

Function: To expel nitrogenous waste products. The excretory fluid is passed on to the intestine through certain canals and ducts, and is ultimately expressed out along with the faeces.

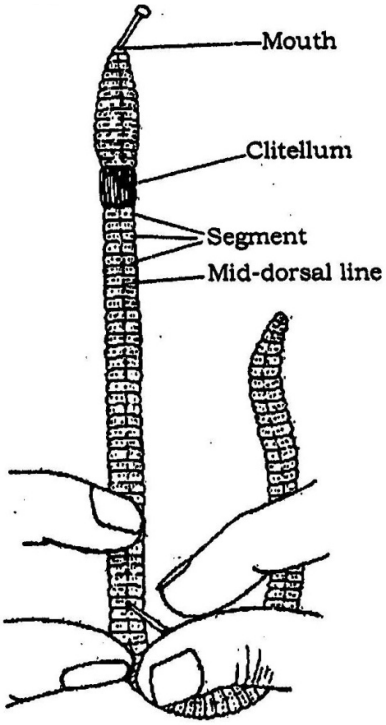


Fig. 10 Pinning the animal

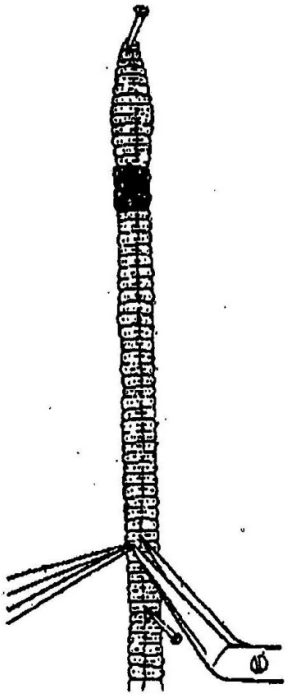
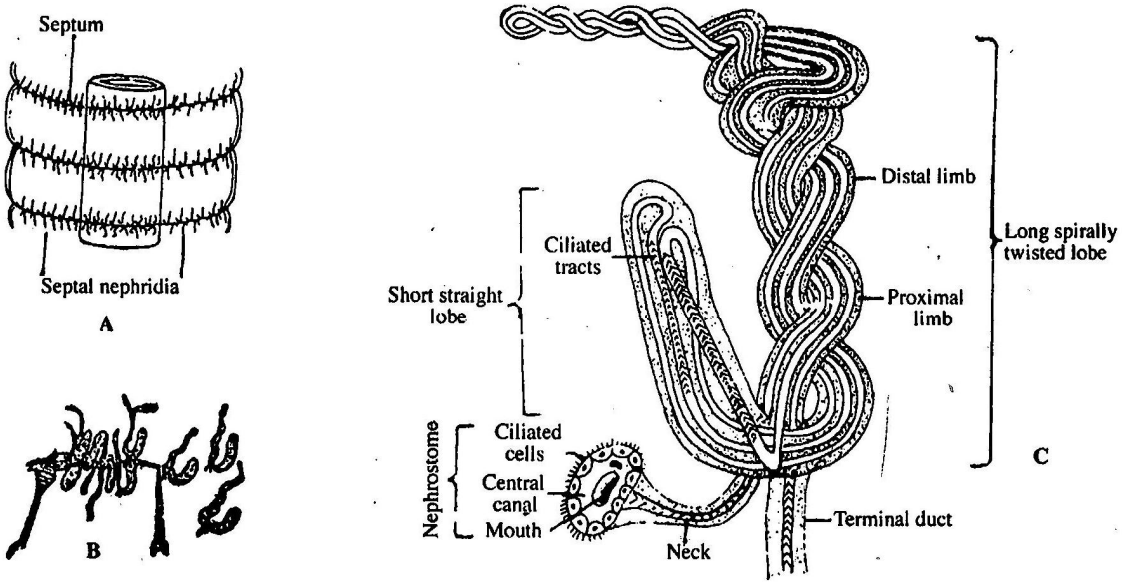


Fig. 11 Cutting the body wall

Fig. 12 Cutting the body wall upto the anterior end



A. Position B. Teasing of nephridia C. Structure
Fig. 21.3 : Earthworm : Septal nephridium

PRACTICAL 5:
MOUNTING OF SPERMATHECA OF EARTHWORM

AIM: To mount and study the spermatheca of Earthworm.

BACKGROUND INFORMATION:

Spermathecae are sperm storing bags placed ventrolaterally. Sperms received from the other earthworm during copulation are stored in the diverticulum while the ampulla contains nourishing fluid for keeping the sperms viable. The narrow neck helps in receiving and shedding of the sperms into the cocoon.

REQUIREMENTS:

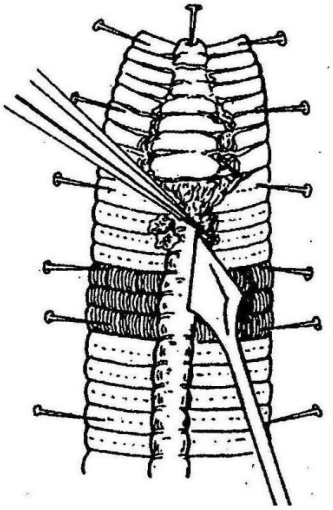
Earthworm, Wax tray, Water, Pins, Forcepsss, Needle, Brush, Hand lens, Filter paper, Eosin, Dissecting microscope, etc.

PROCEDURE:

1. Mark the dorsal side by locating the mid dorsal black line and pin the earthworm in wax tray containing water.
2. Cut open the animal to its anterior region.
3. Loose oblique septa.
4. Remove the alimentary canal so as spermathecae gets exposed.
5. With the help of brush remove the unwanted debris.
6. Remove water by tilting the tray and add new water.
7. Locate spermathecal arrangement in animal with the help of hand lens.
8. Remove spermatheca with Forcepss and place it in watch glass containing eosin.
9. Put stained spermatheca on slide and mount in glycerine.
10. Observe it under dissecting microscope.

OBSERVATIONS AND RESULT:

Spermathecae (singular – spermatheca) are small flask-shaped structures consisting of a pear- shaped ampulla and short narrow diverticulum. There are four pairs of spermathecae occurring along the ventro-lateral sides of the 6^{th}, 7^{th}, 8^{th}, and 9^{th} segements.



Removal of Alimentary canal

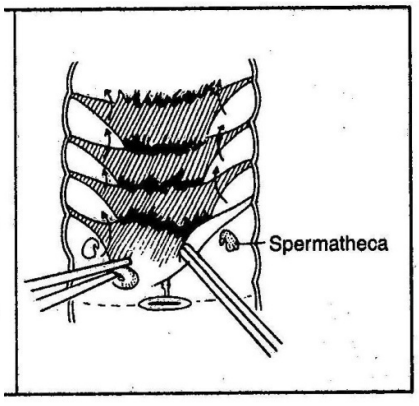


Fig. 22.8 : Tracing the spermathecae

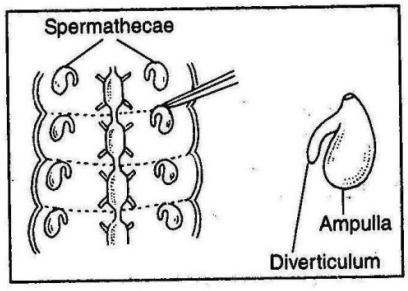


Fig. 22.11 : Removing the spermatheca

PRACTICAL 6:

MOUNTING OF OVARY OF EARTHWORM

AIM: To mount and study Ovary of Earthworm.

BACKGROUND INFORMATION:

Earthworms reproduce sexually not asexually. They are hermaphrodite (monoecious) i.e. both ovaries and testes are found in the same individual. They are protandrous, the male sex cells mature much earlier than the female cells, therefore, self-fertilization is not possible. The relative position of ovary and testes also makes self-fertilization impossible.

REQUIREMENTS:

Earthworm, Wax tray, Forcepsss, Needle, Water, Methylene blue, Glycerine, Filter paper, Compound microscope, etc.

PROCEDURE:

1. Mark the dorsal side by locating the mid dorsal black line and pin the earthworm in wax tray containing water.
2. Cut open the animal to its anterior region.
3. Cut the alimentary canal in front of the clitellum and lift it up to the 12^{th} segment.
4. Locate the ovary lying in between the 12^{th} and 13^{th} segment and pick it up with the help of Forcepsss and place it on slide.
5. Stain it with dilute methylene blue.
6. Remove excess of stain with filter paper and mount in glycerine.
7. Observe it under low power objective of compound microscope.

OBSERVATIONS AND RESULT:

A pair of ovaries remains attached to the posterior face of septum 12/13 one on either side of the nerve cord. They are found in 13th segment. Each ovary is a white compact mass made up of several finger-like processes. The processes contain ova in various stages of development arranged in a linear series.

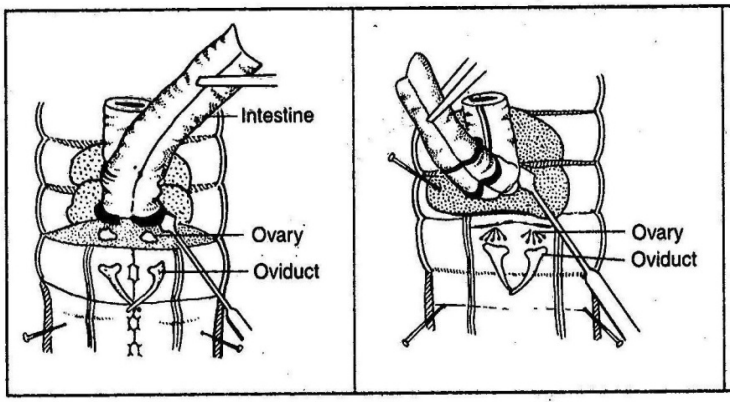
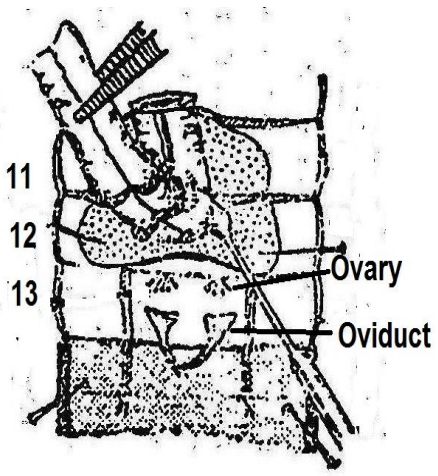


Fig. 22.6 : Separation of alimentary canal for exposing ovary and oviduct

Fig. 22.7 : Tracing the ovaries



PRACTICAL 7:

MOUNTING OF SETAE OF EARTHWORM

AIM: To mount and study Setae of Earthworm.

BACKGROUND INFORMATION:

The locomotion is due to action of muscles in the body wall which are aided by setae. Setae are tiny, bristle-like hairs that extend from most segments.

The setae provide a temporary anchorage to the body on the substratum. When retracted, the setae allow the earthworm to move freely through the soil. Extended setae dig into the soil and hold the worm in place. Earthworms use setae in conjunction with their segment muscles to make burrowing easier. When they contract the muscles in their front segments, they also retract the setae from those segments so the front section of the worm can push forward. The extended setae in the middle and rear sections hold the rest of the body in place. The front ends grab the dirt with setae to hold the segments in place as the middle then the rear segments contract and release their setae to slide forward.

REQUIREMENTS:

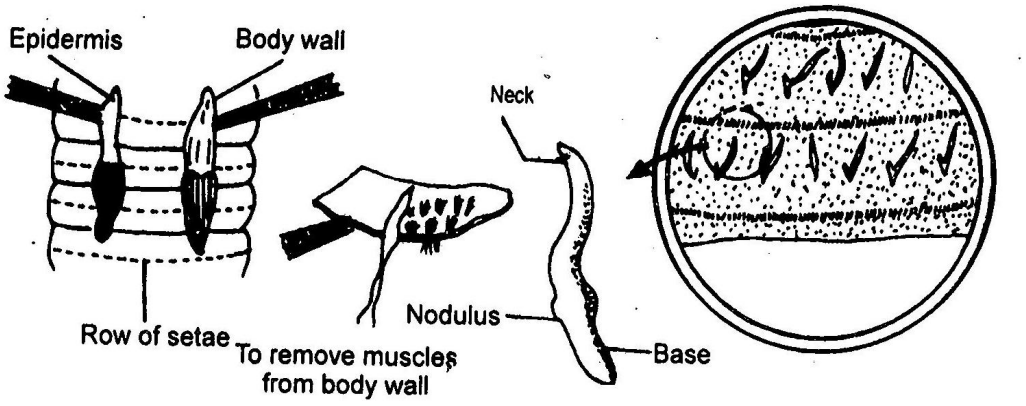
Earthworm, Scissor, Scalpel, Wax tray, Slide, 10 % KOH, Water, Glycerine, test tube, Holder, Burner, Filter paper, Compound microscope, etc.

PROCEDURE:

1. Cut open the earthworm and cut little part of body wall.
2. Transfer it to a test tube containing 10 % KOH solution for removing the muscles from the inner side.
3. With the help of scalpel scrap the epidermis from the body wall.
4. Wash it with water and mount in glycerine on slide.
5. Focus the slide under the high power of compound microscope.

OBSERVATIONS AND RESULT:

Each segment, except the first and last, have tiny bristle-like structures called setae.



PRACTICAL 8:

MOUNTING OF MOUTH PARTS OF MOSQUITO

AIM: To mount and study of Mouth parts of Mosquito.

BACKGROUND INFORMATION:

Mouth parts are the appendages grouped around the mouth. Mosquitoes have mouthparts which are used for piercing and sucking. Typically, both male and female mosquitoes feed on nectar and plant juices, but in many species the mouthparts of the females are adapted for piercing the skin of animal hosts and sucking their blood as ectoparasites. In many species, the female needs to obtain nutrients from a blood meal before she can produce eggs, whereas in many other species, she can produce more eggs after a blood meal. A mosquito has a variety of ways of finding their prey, including chemical, visual, and heat sensors. Both plant materials and blood are useful sources of energy in the form of sugars, and blood also supplies more concentrated nutrients, such as lipids, but the most important function of blood meals is to obtain proteins as materials for egg production.

REQUIREMENTS:

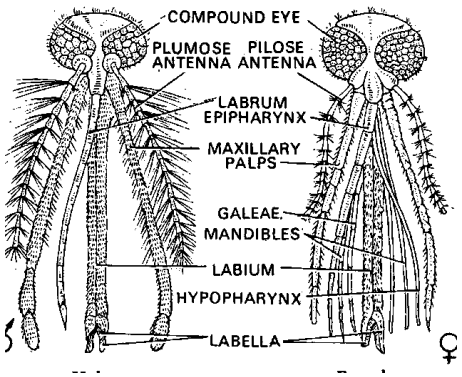
Mosquito, 10 % KOH, Scalpel, Watch glass, Test tube, Holder, Burner, Slides, Filter paper, Dissecting microscope, etc.

PROCEDURE:

1. Cut the head part of Mosquito with the help of Scalpel.
2. Put the removed head in the test tube and add 10% KOH.
3. Attach the holder to the test tube and boil for 2-3 mins on flame.
4. Put the test tube content in the watch glass.
5. Keep the treated head on slide and put another slide on it and gently press so the inner parts will come out.
6. Observe it under dissecting microscope.

OBSERVATIONS AND RESULT:

The paired mandibles and maxillae are formed into needle-like structures (stylets) which are enclosed by the labium. When a mosquito 'bites', the pointed and barbed pair of maxillae penetrate the dermal tissue of the vertebrate to anchor the mouthparts in the tissue. This also provides leverage when the other mouthparts are inserted. The sheath like labium slides back and the remaining mouthparts pass through its tip and into the tissue. The mosquito injects saliva, which contains anticoagulants, into the tissue to stop the blood from clotting. The labrum acts like a tongue and is used to suck up the blood.



PRACTICAL 9:

MOUNTING OF MOUTH PARTS OF HOUSE FLY

AIM: To mount and study the mouth parts of House fly.

BACKGROUND INFORMATION:

Mouth parts are the appendages grouped around the mouth. The housefly is the typical sponging insect. The labium gives the description, being articulate and possessing at its end a sponge-like labellum. The housefly is able to eat solid food by secreting saliva. As the saliva dissolves the food, the solution is then drawn up into the mouth as a liquid.

REQUIREMENTS:

House fly, 10 % KOH, Scalpel, Watch glass, Test tube, Holder, Burner, Slides, Filter paper, Dissecting microscope, etc.

PROCEDURE:

1. Cut the head part of House fly with the help of Scalpel.
2. Put the removed head in the test tube and add 10% KOH.
3. Attach the holder to the test tube and boil for 2-3 mins on flame.
4. Put the test tube content in the watch glass.
5. Keep the treated head on slide and put another slide on it and gently press so the inner parts will come out.
6. Observe it under dissecting microscope.

OBSERVATIONS AND RESULT:

The prominent fleshy and retractile proboscis consists mainly of the labium. The proboscis is grooved on its anterior surface, within this groove lie the labrum-epipharynx (enclosing the food canal) and slender hypopharynx (containing the salivary canal). Mandibles are absent. The maxillae have evidently become fused with the fleshy elbow of proboscis, and only the prominent single segmented maxillary palpi remains. The end of the proboscis is enlarged, sponge like and two-lobed which acts as suction pads. They are called labella. The surfaces of labella are transverse by capillary canals called pseudotracheae, which collect the liquid food and convey it to the food canal. These insects often spit enzyme-containing saliva onto solid foods to liquefy them and then sponge up the mixture.

When labella are pressed against the exposed liquid material, the pseudotracheae absorb it and get filled with by capillary attraction. The liquid is then collected at a point on the labella wherein these tiny channels converge. From this point the liquid is then drawn up through food channel formed in-between the two stylets viz. labrum epipharynx and hypopharynx.