Introduction

This week we conclude the exploration of the phyla within the superphylum Lophotrochozoa. Recall that the name Lophotrochozoa comes from the names of the two major animal groups included: those belonging to Lophophorata and those belonging to Trochozoa. Last week we examined representatives of Lophophorata, the Bryozoans and the Brachiopods. This week we will focus on the two largest groups of Trochozoans, those belonging to the phylum Mollusca and to the phylum Annelida.

These two seemingly different phyla share a fundamental feature of their ontogeny, the trochophore larvae. This larva has two bands of cilia around the middle that are used for swimming and for gathering food, and at the top is a cluster of longer flagellae. So they have a larval stage that is nearly identical even though they mature into very different adult forms. Some Molluscs have evolved additional larval stages that are not found in the annelids.

Molluscs represent the second largest group of animals on earth. According to Campbell Biology, 9th edition, there are 93,000 known species of Molluscs. Molluscs have been extremely important to humans for thousands of years. They have been utilized as a food source, to make dyes, buttons and jewelry, and even fabric. In this lab we will only consider four of the 8 extant classes, Polyplacophora, Gastropoda, Bivalvia, and Cephalopoda.

Annelids are segmented worms that were once thought to be related to the arthropods because of their segmented bodies. However this relationship is not supported by more advanced phylogenetic studies. There are around 12,000 extant species of Annelids and they are found in marine, fresh water, and terrestrial environments.

Although Annelids have not been exploited to the same degree as the Molluscs, they do perform vital roles in the environments within which they are found. Charles Darwin was one of the first to analyze the importance of earthworms to soil fertility. Additionally, marine Annelids are thought increase the penetration of water and oxygen into the sea-floor sediment by burrowing. Finally, a fresh water Annelid, the leech, has been used medicinally for thousands of years. In this lab we will consider three classes of Annelids, Polychaeta, Oligochaeta, and Hirudinea.
Figure 1: Phylogeny of Animalia
Mollusca

Mollusca is the second largest animal phylum after Arthropoda. Molluscs, many of which are familiar shallow marine and fresh water creatures, include clams, mussels, oysters, snails, slugs, octopuses, squid and cuttlefish. They are an important source of food for people and for many other animals. All Molluscs possess two characteristics that are unique to the phylum: a ventral, muscular foot, and the mantle, a dorsal body wall layer that deposits the shell. In many Molluscs, the mantle extends beyond the visceral mass forming the mantle cavity, which is a fluid-filled chamber containing the gills and the anus. Molluscs are triploblastic coelomates that are bilaterally symmetrical. Many possess a chitinous-covered feeding apparatus called a radula used to scrape food from substrates in their environment.

Polyplacophora – Chitons

The chitons (ky-tons) in Class Polyplacophora (“many shell bearer”) show most of the molluscan characteristics in essentially the ancestral condition. The most obvious exception is the shell, which is divided into eight individual valves or plates. These plates are attached to one another via stout ligaments and muscles yet they articulate freely enough to allow the chiton to conform to the irregularities of their rocky substrate habitats. Indeed many of the chitons can roll up like a pill-bug if they are detached from the rocks, a behavior that gives them their other common name, the sea cradles. The chiton creeps along on its broad muscular foot, much like ancestral Molluscs are thought to move. When at rest the foot can obtain such a strong hold on the substratum that is quite surprising.

Find the preserved Chitons on display. Notice that the body has bilateral symmetry, and that the 'head' is indistinct (these are both ancestral conditions for the Molluscs). Find the following structures: dorsal shell plates, muscular foot, mouth, anus, mantle, and the pallial groove containing the gills. There are also openings of the reproductive tract (the gonopores) and the excretory tract (the nephridiopores) located in the pallial groove, but they are very small and you shouldn’t be concerned about finding them.

Figure 2: Chiton
Gastropoda – Snails, Slugs, Abalone, Nudibranchs

Gastropoda means "stomach foot." These creatures include snails and slugs, which are marine, fresh water and terrestrial, the terrestrial slugs, and the marine abalone and nudibranchs. The shell of a snail coils either to the animals left or to its right and provides a home into which the soft animal can withdraw. A slime gland in the front of the foot secretes mucus through which the snail glides by muscle contractions of the foot. The digestive system, excretory organ, mantle cavity and gills are housed within the shell. The mouth of a gastropod contains a rasp-like structure called a radula that can be used to scrape algae off rocks or make holes in the shells of other Molluscs. Slugs and nudibranchs have lost the torsion or twisting of a snail's body. The mantle cavity in a slug functions as a lung, and its entrance, a hole into the side of the animal, can be seen to open and close.

Find the living marine snails in the large aquarium. The aquarium glass has not been cleaned, so you should find snails feeding on algae that coat the glass, and you should find "tracks"- evidence of earlier feeding by the snails.

Find the preserved land snails (Helix) and slugs (Limax) on display.

Examine the preserved land snail under the low power magnification of a dissecting microscope. Observe the muscular foot, the coiled shell, and the distinct head with its two pairs of pre-oral tentacles. The longer pair of tentacles has photoreceptors (or "eyes") on the distal ends. Try to find the pneumostome (an opening to the respiratory space, or 'lung') and, if you hold the snail upside down, you may be able to see the mouth and radula. Be able to identify as many of these structures on the preserved specimen.

Find a prepared slide labeled "Patella (or Whelk) trochophore larva w.m.". Be able to identify a trochophore larval stage.

Bivalvia – Clams, mussels, oysters

Class Bivalvia is a group of Molluscs that differ considerably from the ancestral Mollusc. Members of this class have a shell that is divided in two along a dorsal midline, and held together by a hinge with interdigitating teeth and a flexible ligament. The two valves close by contraction of the adductor muscles. The shell encompasses a body that is compressed laterally (from the side) and has an extended dorsoventral axis and a reduced lateral axis. This compression of the body is seen in changing the foot from a broad, flat, creeping structure to a tapering, blade-like structure for digging. The foot no longer supports the visceral mass, which is now laterally compressed and suspended from the dorsal axis of the shell. The two shell plates and their underlying mantle enclose the foot and visceral mass, creating a large mantle cavity space on either side. Besides the foot and visceral mass the bivalve's greatly enlarged ctenidia (combined gill and filter-feeding structure) are also suspended in the mantle cavity, effectively dividing it into a lower incumbent chamber and an upper excurrent chamber. The head is greatly reduced, consisting of only the mouth; the radula is missing.
First, check out the aquariums. Hopefully we will have a couple of bivalves for you to observe. Some bivalves, like the Giant Clam, rely on the photosynthesis of its symbionts for much of their nutrition. Then, find the preserved freshwater mussels (clams) on display. Examine an intact clam. Then examine one of the specimens that have been opened and look for the following items: **dorsal** and **ventral** surfaces, **anterior** and **posterior** ends of the animal, **shells** (valves) and **hinge**, the **adductor muscles**, the **visceral mass** (gut), the **mantle**, the **incurrent** and **excurrent** siphons, the **gills**, and the **anal opening** within the excurrent channel.

Find the preserved razor clams on display. Look for the structures labeled in Figure 3 on this organism.

**Cephalopoda – Nautilus, Cuttlefish, Squid, Octopus**

Cephalopods (head foot) include squid, octopus, chambered nautilus, and cuttlefish and are marine animals. The **mantle** forms a cavity with a funnel-shaped **siphon**. When the mantle relaxes, the cavity fills with water; contraction of muscle in the mantle squirts out a jet of water. By manipulating the direction of the siphon, the cephalopod can orient its swimming. A threatened octopus can add a dark secretion, **ink**, to the outflow so that it can swim away behind a concealing cloud.

Cephalopods are predators that use **tentacles** with **suction discs** to capture their prey. A **radula** moves the prey to the beaklike jaws of the mouth that bite and crush it. Some cephalopods also secrete venom into the prey. The high levels of activity incur a great demand for oxygen. The circulatory system is closed and highly specialized to pump blood through the gills. At rest a cephalopod rhythmically relaxes and contracts the mantle cavity to facilitate gas exchange in the gills and to expel waste that is discharged into the mantle cavity. The nervous system is highly developed, and cephalopods are capable of learning. They also have excellent vision with **eyes** that are in some ways similar to mammalian eyes. Despite the fact that they are color-blind, some cephalopods are capable of amazing color change. They can match their surroundings so well that they are virtually impossible to see.
Within their skin there are layers of pigment cells, called chromatophores that contain sacs of pigment, with each cell being connected to a number of muscles. When the muscles contract they stretch the chromatophores, spreading out the pigment sacs. As each sac is stretched out the pigment becomes more and more visible. They are also able to match the texture of the substrate against which they are hiding.

Find the preserved squids on display. On these intact animals, locate the dorsal, ventral, anterior and posterior surfaces. Also, identify the tentacles and arms, the eyes, the tip of the pen, the fins, and siphon on the preserved specimen.

Find the preserved octopus on display. Find the same structures as on the squid and compare the two animals. What differences in lifestyle can be inferred from these differences?

Find an instructor to see the cephalopod (Nautilus) shell on display. Look for the siphuncle (connections between chambers).

Annelida

Annelids are segmented worms that live in marine and fresh water as well as in moist terrestrial habitats; some are parasitic. They are bilaterally symmetrical, have a true coelom, and a segmented body. The cellular layer that lines both sides of the coelom is called peritoneum. The digestive system begins at the mouth at one end and runs to the anus at the other. The nervous system has a brain and the blood vessels are closed tubes. Many have gills for respiration, while others (e.g., earthworms) have no specialized respiratory structures. There are no hard parts and the segmented worms are said to have a hydrostatic skeletal support.

Polychaeta – Bristle worms

Polychaetes (many bristles) live in the marine environment and are very common in the shallow, intertidal zone. Many are carnivores and prey on a variety of invertebrates. Nereis has pinching jaws with horny coverings with short sensory palps located immediately behind the jaws. Tentacles, or cirri, arise from the segments immediately behind the palps. The lateral appendages are called parapodia and are used for locomotion. The parapodia enable the worm to “row” through the water and to excavate a burrow. They also function in gas exchange as they serve as gills.

Figure 5: Nereis
Find the preserved specimens of *Nereis* on display.

Examine the external morphology of this marine polychaete worm. Be sure you can distinguish the head and tail regions, the individual body segments, and the parapodia. Examine the head to find the two specialized pre-segmental units, the *prostomium* and the *peristomium*. The first of the metameric segments, bearing parapodia, is found right behind the peristomium. The prostomium bears both prostomial palps and tentacles. The peristomium has only peristomial tentacles. You should be able to find very small eyes on the upper surface of the prostomium. The pharynx can be extended in *Nereis*.

Find a prepared slide labeled "*Nereis* parapodium w.m."

Parapodia work for both movement and respiratory function. How?

Find a slide labeled "*Nereis* c.s."

Notice the longitudinal and circular trunk muscles, the *ventral nerve cord*, the *coelom* and the intestine with its finger-like projections into the lumen. We have had a few polychaetes in the large aquarium. The Bubble Anemone has periodically covered up the tubes in which they have resided, so they may no longer be alive. Try to find the polychaetes we have in the aquariums. Ask for help if you cannot locate them.

**Oligochaeta – Earthworms**

The name refers to the few (oligo-) bristles (-chaeta) along the body of these earthworms and fresh water worms. Examine the earthworm genus *Lumbricus*. The dorsal or top is slightly darker than the ventral side. Earthworms burrow through soil and eat decomposing plant and other organic material. The mouth end is pointed and conical, and the tail end is dorsoventrally flat. The anus is a vertical slit in the last segment. As you run your fingers along the sides of the living worm, you can feel the rasp of the setae (singular seta) or bristles. Tiny excretory pores from the nephridia can be found on the lateral or ventral surfaces of all segments except those at the ends. The earthworm has both ovaries and testes. The openings of the oviducts can be seen on the sides of segment 14 and those of the sperm ducts, which have swollen edges on segment 15. The enlarged ring that begins at segment 31 or 32 and ends at 37 is called the clitellum. It is glandular and secretes a slimy mucus around two copulating individuals.

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*Figure 6: Lumbricus*
Find a prepared slide labeled “Lumbricus 3 regions c.s.”

Examine the right hand section (as you look through the microscope). Observe the longitudinal and circular muscles, the large coelum, the ventral nerve cord, and the intestine.

Hirudinea – Leeches

Hirudinea simply means leech. Leeches live in both marine and fresh water and in damp terrestrial habitats. The body is segmented, but this is not clearly visible on the outside. A sucker at each end is used for locomotion on a surface; swimming is achieved by contractions of the body wall musculature. Leeches lack setae and parapodia. Some are predators while others are parasites. Those that feed on blood have sharp jaws and secrete an anticoagulant that prevents clotting of the blood. Medicinal leeches are a fresh water variety that is still in use.

Find the preserved leeches on display. (And see Figure 7)

Observe the mouth, suckers, false segmentation and eyes. Unfortunately we will not have living leeches to examine this week.

Figure 7: Leech