Biology 337 *NAME:* ________________________ **Lab worksheet**
Biology of Invertebrates *LAB TIME:* ____________________ **Dr. Bob Podolsky**

LAB G. Ph. ARTHROPODA, Subph. CRUSTACEA, Cl. Malacostraca & Cl. Maxillopoda

Arthropods dominate the planet. Their diversity at every taxonomic level is remarkable. Here we will examine members of Subph. Crustacea. In a later lab we will examine mostly terrestrial representatives in the subphyla Tracheata and Chelicerata, and in the final lab we will come back to look at smaller-bodied, planktonic crustaceans that are members of the Cl. Maxillopoda.

Also remarkable is the fact that *different* arthropod taxa have come to dominate different parts of the biosphere. Crustaceans are the major portion of animal biomass and diversity in oceans, although some are found on land (e.g., isopods and land crabs). Tracheata and Chelicerata are the major portion of animal biomass and diversity on land, although some chelicerates are marine (e.g. horseshoe crabs, pycnogonids) and some terrestrial insects have freshwater larvae.

Crustaceans are a major component of both planktonic and benthic marine communities. All crustaceans have at least some biramous appendages and two pairs of antennae. Many have a **nauplius** larva during their development, although other larval types have evolved. Most incorporate mineral salts (e.g. $CaCO_3$) into the exoskeleton, which is made of sclerotized cuticle (the carbohydrate polymer chitin and proteins).

>>>**General goals**. This lab will include a great deal of diversity in the crustacean body plan. Your goal is to focus on traits that distinguish classes and orders: shape and organization of **tagmata**; shapes and functions of **appendages**; and methods of **feeding** and **movement**.

A. TAXONOMY. Crustaceans are one of three extant subphyla (along with an extinct subphylum, the Trilobitomorpha), recently recognized as a sister group to the **Tracheata**, together comprising the group **Mandibulata**. The third subphylum, **Chelicerata**, is sister to the Mandibulata. A complete crustacean taxonomy is extensive—you should learn the taxonomy to the extent given below in **bold**:

Cl. Malacostraca – About half of all crustacean diversity, this class includes most of the familiar groups (described below). Primitive characters include a carapace, stalked eyes, and a biramous first antenna. Most have 19 body segments (5 head, 8 thorax, 6 abdomen) with one pair of appendages per segment. ("Primitive" body plans have a 7^{th} abdominal segment.)

- **Supero. Eucarida:** carapace fused to all 8 thoracic segments, no brood pouch
  - **O. Decapoda** – includes many infraorders and looser categories: "natantians" (shrimp), "macrurans" (lobsters and crayfish), Infrao. Brachyura (crabs), Infrao. Anomura (hermit crabs and relatives), Infrao. Thalassinida (mud shrimp and ghost shrimp). *Natantians and macrurans are now recognized as grades that are combined from several infraorders.*
  - O. Euphausidacea – includes pelagic krill, major grazers in colder oceans
- **Supero. Peracarida:** carapace fused to no more than 4 segments; **oostegites** form brood pouch
  - **O. Isopoda** – marine isopods, terrestrial pillbugs
  - **O. Amphipoda** – gammarids (beach fleas), caprellids (skeleton shrimp), hyperiids
- Supero. Hoplocarida: the carapace is fused to 3 segments
  - O. Stomatopoda – stomatopods = "mantis shrimp"

Cl. Maxillopoda

Subcl. Cirripedia – Thoracican barnacles glue themselves, head-first, onto hard substrata, produce a set of external calcareous plates, and filter-feed using cirri. Acrothoracicans bore into calcareous structures but also filter feed. Related groups (rhizocephalans and ascothoracidans) produce no plates and are parasitic.

Subcl. Copepoda – Small and typically free living (though some parasitic forms exist), dominate planktonic marine communities and are primary grazers of phytoplankton (some are predatory)

Subcl. Ostracoda – Typically small, bivalved with a symmetrical shell. Most have simple eyes, though one group does have compound eyes (like most other crustaceans). These animals have the distinction of having the largest sperm in the animal kingdom!

Subcl. Branchiopoda – Small, free-living, dominate freshwater planktonic communities (though some marine). Include brine shrimp (sea monkeys) and water fleas (*Daphnia*).

B. Exercises

We start with brachyuran decapods because your dissection of a blue crab will be a starting point for comparison with other decapods and more distant relatives. Note, however, that the crab body plan is *unusual* among malacostracans, with a broad carapace and a highly reduced abdomen.

Cl. Malacostraca, Supero. Eucarida

O. Decapoda: All have a prominent carapace, three pairs of maxillipeds, and five remaining pairs of thoracic pereopods (ten "legs", the name of the order). Usually the first pereopodis are chelate (called a "cheliped" if enlarged, as in crabs). Most brood embryos on the abdomen, holding them with the pleopods. We will examine five groups of decapod crustaceans.

***1. Brachyurans** ("short-tail")* — the "true" crabs

- <u>Movement</u>. If a live blue crab is available in an aquarium, watch the coordinated movements of the legs as it swims and walks. If the aquarium is large enough, disturb the animal to see it swim, using the paddle-like fifth pereopods for rapid movement off the surface. Observe the first pair of pereopods, the **chelipeds**, used for cutting and crushing. Watch the flicking and grooming of the first antennae (located between the stalked, compound eyes). The antennae are covered with tiny sensory hairs that are used to pick up chemical cues from the water. The animal will constantly groom the antennae and mouth area to keep them clean.

TQ: Why do the antennae of crabs flicker? *[Hint: What do these structures lack that would be used for the same purpose in other phyla?]*

- Dissection. Carry out a dissection of a live blue crab. The animal will have been anaesthetized in carbonated water, which uses CO_2 to knock out the animal but doesn't necessarily kill it, so you may still see a beating heart. You will use this detailed look to begin your comparison of characteristics of other crabs and more distantly related crustaceans.

Dissection-TQs (questions found on dissection handout)

DTQ: How does the "serial homology" among articles differ from previous definitions of homology used in this course? (*Hint: both involve the concept of non-independent evolution*.)

DTQ: Given the presence of this special membrane to limit blood loss, why have the basis and ischium become fused? (*Hint: aside from an articulating membrane connecting two plates, what do joints need?)*

DTQ: Given its location, what do you think might be the function of the flabellum? How would it be operated, and why would its operation compromise another function?

DTQ: Cutting into the carapace caused a large amount of fluid to spill out. Transfer some onto the countertop—does it appear to have any color? What is this fluid? Where did it come from?

DTQ: What structure does a crustacean use to generate water flow through the branchial chamber? How does this mechanism differ from the mechanism for water flow through a bivalve mantle cavity? How is it similar to water flow through a cephalopod mantle cavity?

DTQ: Given what you know about the path of water flow and counter-current gas exchange, predict whether you are seeing the afferent or efferent vessel at the top of the gill in your dissection and explain why.

- Molt. As in all arthropods, to grow larger the animal must molt the old exoskeleton, produce a soft new exoskeleton, inflate the body by taking up water (or air, in terrestrial species), and then actively **sclerotize** and deposit minerals in the soft cuticle. The specimen on display of *Portunus* (from the same family as blue crabs—note the paddle-like 5^{th} pereopod) was preserved just after its molt, showing the recently-molted exoskeleton and the soft new cuticle.

Brachyuran comparisons. Now compare portunid crabs to those from several other brachyuran families on display. Your goal is to appreciate the variety of evolutionary pathways that have been followed in terms of the prominence of **tagmata**, **carapace shape**, and **appendage design**, and how these are related to **habitat**. Do not draw every crab whole—you should focus on features that show large differences.

- Shore crabs. This large group of crabs (e.g. *Hemigrapsus* spp., F. Grapsidae) is extremely common in near-shore habitats. These small animals were collected from floating docks. Disturb them to note differences in their movement from the swimming Portunids.
- Golden crab. The deep sea crab *Geryon fenneri* (F. Geryonidae) is especially large for a brachyuran (note that the largest crustaceans, the king crabs, are actually anomurans, related to hermit crabs.)
- Ghost crabs. Like many crabs, the Atlantic ghost crab *Ocypode quadrata* (F. Ocypodidae) feeds at night and remains burrowed during the day. Burrows of young crabs are found closer to shore and those of older crabs are farther from water. The burrows of ghost crabs are common at Folly Beach, especially near the wooden walls at the northeast end. Note the square carapace that gives the species its Latin specific epithet *quadrata*.

- Fiddler crabs. Another member of the F. Ocypodidae, these crabs are also semi-terrestrial. Among differences from other crabs, note that some of the animals exhibit enormous size asymmetry in the chelipeds.

2. Anomurans — ("anomalous tail")

This heterogeneous group has one major shared characteristic: the **fifth pair of pereopods** (and sometimes the fourth) is always highly reduced in size. This pair typically sticks up into the branchial chambers and is used to clean gills or hold onto the inside of a shell (in hermit crabs).

- Hermit crabs (live). Hermit crabs have a soft and flexible abdomen that is tucked into a hard structure produced and abandoned by another animal, usually a gastropod mollusc (though sometimes a polychaete tube). Hermit crabs will frequently exchange shells and individual crabs will try out new shells.
- Hermit crabs (preserved). The preserved specimen out of its shell shows the soft and asymmetrical abdomen, noticeably different from the brachyurans. Find the final two pairs of pereopods, and sketch their shape and size relative to the other pereopods and relative to the homologous stuctures in brachyurans.
- Hermit crabs (molt). A dried molt of a hermit crab shows how much of the body surface must be covered by new cuticle at the time of molting. Note that each leg is just a thin-walled cylinder with lots of empty space inside into which the soft tissue of the animal grew before it molted. In many respects, it is similar in design to the thin, hollow tubes used to make bicycle frames.

TQ: Imagine concentrating the same amount of cuticle (or bicycle frame) into a narrow rod (like an endoskeleton bone) instead of a hollow cylinder. Which design would be better at resisting bending forces? Why? (*Hint: it might help to draw a diagram of the two kinds of skeletal rods bending to the same curvature.)*

- "Squat lobster". Note: not all anomurans are hermit crabs! The so-called squat lobster, another anomuran, is more closely related to hermit crabs than to lobsters. As with all anomurans, count the appendages to identify the **highly reduced fifth pair of pereopods**.

3. "Natantians" — The "shrimp" body form is common to several infraorders, formerly grouped into the Natantia. The body form is now recognized as a grade found among several clades. The examples here are all from the Infraorder Caridea, the most diverse of the shrimp-form clades.

- Glass shrimp (live). Live individuals of *Palaemonetes* will allow you to examine the use of different appendages for three types of shrimp locomotion: **walking**, **swimming**, and **escape**. In a cartoon sketch of shrimp, indicate which appendages are used in each activity (including the direction of limb and body movement), and show how the form of each appendage relates to its use.

4. "Macrurans" — The "lobster" body form, with a heavier thorax and shorter abdomen than shrimp, is also common to several infraorders, formerly grouped into the Macrura. The stouter body form is associated with bottom-dwelling and crawling (rather than swimming) animals. Many live stationed in burrows or cavities.

- <u>Crayfish</u>. Crayfish and true lobsters are in the order Astacidea. Examine a preserved male and female of *Cambarus sciotensis*, the common local crayfish. Note the large abdomen with small **pleopods**, which cannot be used for swimming, as well as the two pairs of large **uropods** that form a tail fan. Examine the pleopods carefully: just as in the crabs, the female **pleopods** are used to hold eggs, and the male first **pleopods** are used in sperm transfer.
  If you examine carefully the ventral surface of the bases of the second antennae, you can locate the openings of the excretory "**green glands**" that are used for osmoregulation in fresh water habitats (one of the only coelomic compartments in crustaceans).

5. Thalassinidea — Ghost shrimp and mud shrimp are champion burrowers, often living in a Y or U shaped burrow in shallow water mudflats. Focus on changes in body shape and how they relate to **body shape**, the cand organ, particularly how segments are organized into tagmata, and appendage shape, particularly Note differences in body shape and appendage shapes when compared with other decapods—consider how these differences relate to their burrowing habits.

CL. MALACOSTRACA, SUPERO. PERACARIDA

As distinguished from the Eucarids, members of this superorder (1) lack a carapace, and (2) brood young in a ventral, *thoracic* brood chamber called the **marsupium** (recall that, in contrast, many eucarids brood on the abdomen). The bottom floor of the marsupium is created by **epipods** (extra branches) of the female's thoracic appendages with the special name of **oostegites**. The young develop in the marsupium all the way until they are fully formed juveniles (in most cases). Therefore, unlike most crustaceans, peracarids (3) have "direct development" that does not include a larval stage. Only two of the five orders are on display.

O. Isopoda

- <u>Giant isopod</u>. We are fortunate to have a preserved specimen of *Bathynomus giganteus*, the largest isopod species in the world (and one of the largest crustaceans), found at depths of more than 2000 ft. As you examine species of more typical size, marvel at how conserved the basic body form is despite the range of sizes. Take advantage of this specimen to be sure you can identify the ***tagmata*** of an isopod: **cephalon** (what does this mean?), **thorax** (and its 7 visible segments), and **abdomen** (and its 6 visible segments) — as well as each of the ***appendages*** of an isopod: 2 **antenna**, 3 **mouthparts** (not visible), 1 **maxilliped**, 7 **pereopods**, 5 **pleopods**, 1 **uropod**, all preceding the terminal **telson**. Note the form of the appendages, and try to guess whether this specimen is male or female depending on the presence or absence of **oostegites**.

- <u>Live isopods</u>. If live animals are available, pick one up (it won't bite!) and take it in a bowl back to your bench —try to get at least one female. These are easy to handle, as they demonstrate "thigmotaxis" (a tendency to move toward and cling to surfaces). Note the **D-V flattening** of the body. Note two types of locomotion—walking and swimming, and again relate these activities to the appendages used. It is possible that some of the females will be brooding embryos. If so, note the **marsupium** created by the **oostegites** (projections from the thoracic legs). Examine any embryos or more developed young under higher magnification to note that these animals undergo **direct development** without a larval stage.

O. Amphipoda

Like isopods, amphipods are abundant small crustaceans in fresh and salt water habitats. Although some occupy the zone just above the high-tide mark in beach habitats, none are as terrestrial as the isopod pillbugs. An important difference in body design is that amphipods tend to be L-R compressed, unlike the D-V flattening of most isopods.

- Gammarid amphipods. These animals have the "typical" amphipod body form, with a laterally-compressed, stout body. If living specimens are available, observe their style of locomotion under a dissecting microscope, and compare this movement to that of isopods. Some specimens may be in amplexus, with the *male* carrying the *female* in anticipation of her molting (the time when mating will occur). Some may be carrying a brood in a brood pouch on the thorax.

  If there is time, place a living specimen on a slide and examine it using a compound microscope. If the specimen is clear enough, identify the **gut**, beating **heart**, and, if it is a female, the **ovaries** (brightly pigmented in mature females and lying along the gut). Examine a leg closely to see the blood circulating. Note the first two pereopods, which are **subchelate** as in all amphipods.

- Caprellid amphipods. So-called "skeleton shrimp" are known for their elongate body form. These animals typically cling to colonies of hydroids, algae, or sponges, with the body extended and swaying to find the next surface to crawl on. The **oostegites** that make up the floor of the **marsupium** are especially clear in females. Make sure you locate and compare at least one male and one female. Are any of the females brooding embryos?

TQ: Compare the major type of movement by (a) gammarid amphipods, (b) caprellid amphipods, and (c) isopods. Which appendages contribute most to movement of each?

- Hyperiid amphipods. These unusual animals are rumored to be the model for the creature in the movie Aliens. They are all planktonic and typically live in association with larger gelatinous animals (jellyfish or salps). In fact, the hyperiid genus Phronima specializes on kicking out its host (a salp, which has a clear outer house), taking over the house, and spending its life propelling itself through the water by kicking water through the house with its pleopods!

CL. MALACOSTRACA, SUPERO. HOPLOCARIDA
O. Stomatopoda

- Mantis shrimp. The common name, "thumb splitter", reflects the ability of these animals to jab with their large subchelate claws and sharp uropods. Some species are "smashers" that can use appendages to thump and crack open prey (and, unfortunately aquarium glass!). If preserved stomatopods are on display, note the large, well developed **compound eyes** and the large, **subchelate claws** used for capturing prey, such as shrimp and fish. Note also the **flat carapace** (fused only to horacic segments 1-3, unlike in Eucarids where it is fused to all 8) and the large **abdominal pleopods**. Identify the tail fan made up of **uropods** on either side of a large telson.

CL. MAXILLOPODA, O. CIRRIPEDIA

The only taxon in the maxillopoda that you will examine today is barnacles. Begin by examining a rock surface coated with a number of settled balanomorph thoracican barnacles. Recall that thoracicans are hermaphrodites and can mate only with nearby individuals.

TQ: The barnacle penis is the longest for its body size among animals. Devise a scaling argument for how the number of mates is expected to change as a function of body volume, and therefore how you would expect penis size to scale with body size. (Use mathematical notation and a simple diagram.)

- Inspect the inside of the empty barnacle shells. Note that the shell consists of six, adjoining plates. In order to grow, the barnacle must secrete new plate material, like a mollusc. Note that plates must stay attached to each other and to the substrate even as they grow larger—how can a barnacle achieve this? Where is new plate material being added?
- Watch live barnacles, if present, use the modified thoracic legs (cirri) to filter suspended food. Note that because barnacles filter particles using a capture area, as their body

TQ: Based on a scaling relationship, predict how the length of cirri are expected to change as a barnacle grows in size. (Use mathematical notation.)