

## Biology 3B Laboratory

### Protist Diversity

#### Objectives

- Learn the basic characteristics that define organisms classified within the Protist taxon
- To learn the anatomy, life cycles and identification of representative organisms from the principle Protist phyla
- To demonstrate an understanding of the ecological and economic importance of organisms within this taxon

#### Introduction

Within the Domain Eukarya, Campbell and Reece erect eleven kingdoms (Fig. 1). Three of these are the Fungi, the Animals and the true Plants; the balance are a hodge-podge and diverse collection of eukaryotes. This eight kingdom taxon has historically been referred to as the Protista. Most of the members are single-celled organisms (originally called the protozoa), but also included are colonial species and true multi-cellular algae (these used to be included among the plants). Within these eight kingdoms, Campbell and Reece recognize 16 Phyla.

Two kingdoms, the Diplomonads (Diplomonadida) and Trichomonads (Parabasala), are generally considered pleisiomorphic to the greater bulk of protistans. Although they lack mitochondria, this is not a primitive character; rather it is fairly well accepted that these organism possessed and subsequently lost these cytoplasmic symbionts. You're in luck, as we will not look further at these two groups.

Our first major group will be the Euglenozoa. Within this kingdom there are two phyla, Euglenophyta and Kinetoplastida; both of these possess **flagella**. In this lab you will carefully study living and prepared *Euglena*. The only Kinoplastid you must know is *Trypanosoma*, the organism that causes African sleeping sickness.

The Kingdom Alveolata is large and diverse (from the perspective of a biology student). There are three phyla within this taxon: dinoflagellates (Dinoflagellata), apicomplexans (Apicomplexa) and ciliates (Ciliophora). Dinoflagellates are single-celled protists found in marine and fresh water ecosystems; they comprise an important part of the phytoplankton. For example, *Gonyaulax* is a local dinoflagellate responsible for **red tide** along our coast. The common apicomplexan that you must know is *Plasmodium*, the organism responsible for **malaria** in humans (see figure 28.13 in Campbell and Reece). The Ciliophora, as the name suggests, use **cilia** to move and feed. In the lab you will see live *Paramecium*, *Blepharisma* and *Stentor*.

Water molds (Oomycota) and several types of **flagellated algae** are grouped into the Kingdom Stramenopila. The water molds are all **heterotrophic**, most are **single-celled**, and some are made up of branching **coenocytic hyphae** (analogous to hyphae of the fungi). There are several parasitic water molds of economic

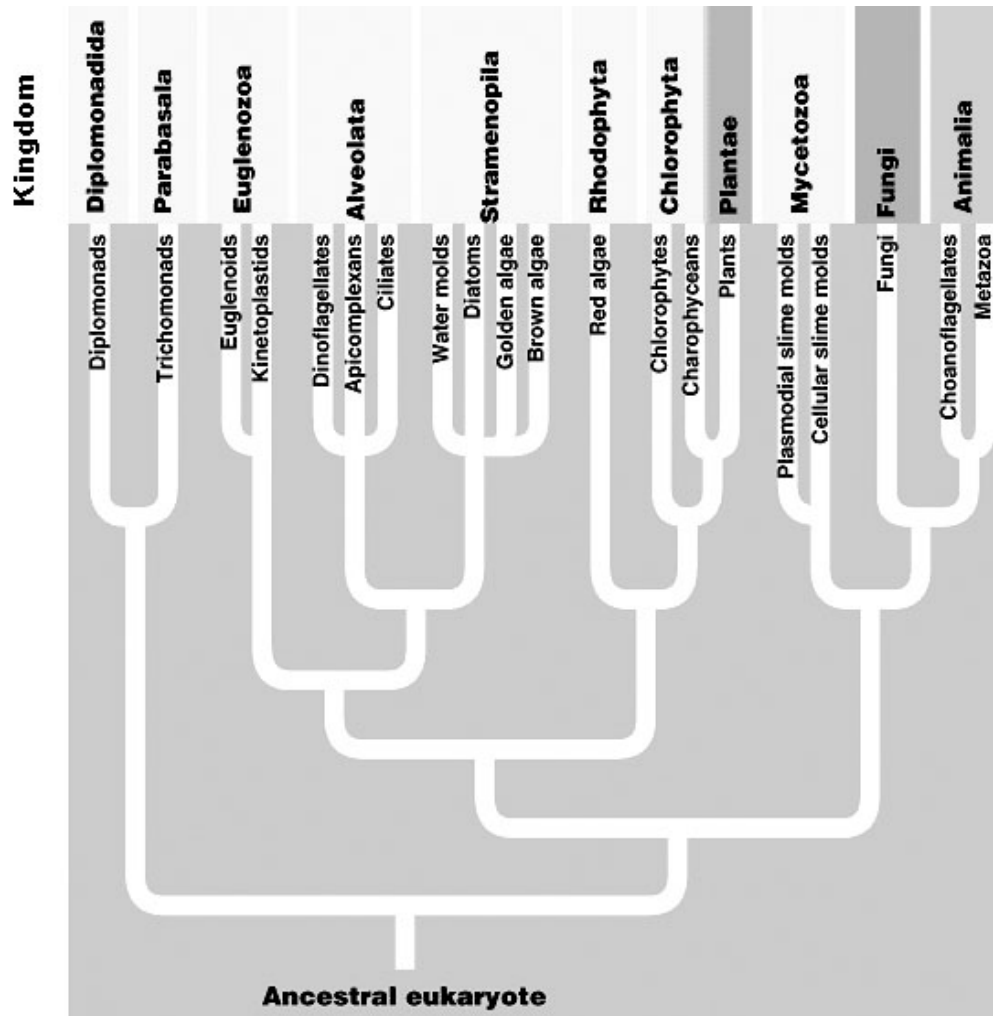
importance including downy mildew (*Plasmopara viticola*) that attacks grapes, *Phytophthora infestans* that causes potato blight, and *Saprolegnia* that causes lesions on fish. The algal phyla within the Stramenopila are the diatoms (Bacillariophyta), golden algae (Chrysophyta) and brown algae (Phaeophyta). Diatoms comprise another important part of the phytoplankton of marine and fresh water ecosystems. They are single-celled organisms that have a **silica** (siliceous) skeleton consisting of two overlapping halves (not unlike the two parts of a Petri dish). Golden algae are **biflagellated, mixotrophic** marine and freshwater organisms. In addition to photosynthesis, they may feed by absorption of chemical compounds and phagocytosis. The brown algae are a **very large, multicellular**, mostly marine group. Some of these may grow to incredible size (e.g. *Macrocystis spp.*, the giant kelp).

There are two other algal kingdoms within the protists. The Rhodophyta are the red algae. The red color derives from a pigment called **phycoerythrin**. The plastids containing this pigment are believed to have originated as cyanobacteria incorporated into the red algae by **primary endosymbiosis**. The Kingdom Chlorophyta are the green algae. As the name suggests, these organisms have chloroplasts and are probably closely related to the true plants. It is the cells of chlorophytes that live symbiotically with fungi to create the “organisms” we call lichens. In this lab you will observe live *Volvox globator*, a colonial chlorophyte.

The two taxa of slime molds (Myxogastriada and Dictyostelida) are primarily decomposers. These diverse organisms may live as one-celled amoebas and at other times aggregate into multi-celled masses more than a meter across. Most are saprophytic, feeding on dead plant or animal remains, but some are parasitic.

Finally we are left with three phyla of uncertain affinities. Campbell and Reece find some value in grouping these by their use of **pseudopodia** for locomotion and feeding. The Phylum Rhizopoda are the amoebas. In lab you will examine living specimens as they move and feed by extension of pseudopodia. The phylum Actinopoda are aquatic organisms with siliceous skeletons. You will look at the skeletons of radiolarians in lab. Finally, the Phylum Foraminifera are a diverse group of mostly marine protists. Many are abundant in plankton, while others are found attached to rocky substrates or within benthic sediments. The shells of these organisms is usually multi-chambered, composed of **calcium carbonate**. Each of the chambers is connected to the others by a hole, or foramen. Thus each animal has several foraminifera.

As you examine the diverse samples notice how this taxon ranges from single-celled to multicellular, from heterotroph to autotroph, and from structurally simple (single celled) to complex. Look for characteristics that are shared with animals, plants and fungi; most evolutionary biologists believe the remaining multicellular kingdoms arose from protist ancestors.



**Figure One.** Proposed phylogeny of the Eukaryotes

### Procedure

There is a large amount of material to cover in this lab section. My approach will be phylogenetic; that is I will begin with the first group on Table 1 and proceed toward the last. For each organism you should make careful observations on physical makeup and behavior (for living organisms). Important features to include: size, general appearance (careful drawings are important), internal organization, movement, feeding behavior, and reproduction. You will not be able to observe all of these characteristics in lab. Missing information may be found in your textbook and on the internet. When these observations are complete and organized, you can then compare and contrast different groups within the Protista. Our ultimate goal is to present a clear statement of defining the taxon Protista.

### Kingdom Parabasala and Kingdom Diplomonadida

You will be happy to know that we have no slides of these organisms (at least that I could find)! Read about them in your textbook and move on to the next kingdom.

## **Kingdom Euglenozoa**

### **Phylum Euglenophyta**

Examine both live and whole mount slides (w.m.) of *Euglena*. If you do not remember how to prepare a wet mount (of the live organisms) please ask for a demonstration!

On the slides and live specimens find the (and know the function of)

**nucleus**  
**paramylon bodies**  
**cytopharynx**  
**flagellum**  
**eyespot**

Observe the movement of a live *Euglena*. Using the eyepiece scale (reticule) and a slide micrometer make some measurements of the total length of several *Euglena*. You may need to add some methylcellulose to slow it down. Carefully draw a series of pictures to illustrate this “euglenoid” movement. Write a short description of the locomotion of this organism.

## **Kingdom Alveolata**

### **Phylum Dinoflagellata**

Examine prepared slides of *Noctiluca* and *Ceratium* and find

**flagellar groove**  
**flagellum**

### **Phylum Ciliophora**

Examine *Paramecium*, *Blepharisma*, and *Stentor* both alive and on prepared slides. Try to watch feeding in one of these organisms. Be sure to find

**cilia**  
**macronuclei**  
**micronuclei**  
**food vacuoles**  
**oral groove (*Paramecium*)**

Use a few *Paramecia* or *Stentor* to perform the following test. Place them in a depression slide and observe under a dissecting scope. Now place a drop of vinegar or salt water next to the depression. Using a toothpick connect the drop to the solution in the depression. What is the reaction of the organisms to each of these solutions?

## **Kingdom Stramenopila**

### **Phylum Oomycota**

Look at the leaf section of a potato infected with *Phytophthora infestans*. On the under side of the leaf look for red sporangioophores penetrating the stomata. You may also see oval sporangia near these.

Take a quick look at the plants growing around the building. Can you find any examples of water molds? What areas might be the best places to look?

### **Phylum Bacillariophyta (diatoms)**

Examine slides of diatoms. Draw several different types (shapes).

### **Phylum Phaeophyta (brown algae)**

Since these are very large organisms, we can examine herbarium sheets of pressed examples of these organisms. Look at several species of the phaeophytes, including *Macrocystis*. Sketch some of the different morphologies. Next, sketch out the life cycle of a typical brown algae.

### **Kingdom Rhodophyta (red algae)**

These are also large organisms. Look at several species (in the herbarium collection) of the rhodophytes. Sketch some of the different morphologies. Pay close attention to the coralline species. (What is the role of red algae in a tropical coral reef?)

### **Kingdom Chlorophyta (green algae)**

Okay this is a diverse group. First, you must examine the live specimens and prepared slides of *Volvox*. Be sure to sketch the live colonial organism. Find the **daughter colonies**

Now, look in the herbarium sheets for a green alga. What is the relative size of marine green algae?

### **Other Phyla**

#### **Phylum Rhizopoda (amoebas)**

Examine both living specimens and prepared slides of *Amoeba proteus*

You should be able to see

plasmalemma

contractile vacuole

nucleus

food vacuoles

pseudopodia

In the living specimen you should create a series of 6 small sketches, each one showing the outline of a single amoeba about every thirty seconds. Essentially, you are creating a diagram showing locomotion. Using the scale in the eyepiece of the scope and a micrometer microscope slide can you estimate the speed of amoeba locomotion?

Watch a single contractile vacuole for a period of time. It should appear and then disappear. Time the "life" of a contractile vacuole, then, replace the culture

medium on the slide with distilled water. This is done by placing a drop of DI water on one side of the cover slip and placing a piece of absorbent paper on the other, the DI water will be drawn into the depression on the slide. After you have done this, time the “life” of another contractile vacuole. Compare both measurements.

**Phylum Actinopoda** (radiolarians and heliozoans)

Examine prepared slides of radiolarians. Draw at least two shapes of tests.

**Phylum Foraminifera** (foraminiferans)

Examine prepared slides of foraminifera’s. Look closely at the tests. From what material are the tests constructed? Can you see the foraminifera?

**Table One.** Classification Scheme for the “Protistan” Eukarya (based on Campbell and Reece 2002.

**Domain Eukarya**

**Kingdom Parabasala** (trichomonads)

Lacking mitochondria

*Trichomonas vaginalis*, vaginal and urethral parasite

**Kingdom Diplomonadida** (diplomonads)

Two separate nuclei, no plastids, no mitochondria, multiple flagella

*Giardia lamblia*, human intestinal parasite

**Kingdom Euglenozoa** (euglenoids and kinetoplastids)

Flagellated, may be autotrophic, mixotrophic or heterotrophic

Phylum Euglenophyta

*Euglena*, organism seen in live in this lab

Phylum Kinetoplastida

*Trypanosoma*, causes African sleeping sickness

**Kingdom Alveolata** (dinoflagellates, apicomplexans, ciliates)

Membrane-bound alveoli under cell surface

Phylum Dinoflagellata (dinoflagellates)

Microscopic algae that form the basis of most marine food chains

*Gonyaulax*, responsible for red tides

Phylum Apicomplexa

Parasites of animals

*Plasmodium*, causes malaria in humans

Phylum Ciliophora (ciliates)

Move by undulating cilia, feed by ingesting bacteria or other protists, marine or freshwater

Paramecium, Stentor, Blepharisma

**Kingdom Stramenopila**

Phylum Oomycota (water molds, powdery or downy mildew)

unicellular or coenocytic

*Phytophthora infestans* causes potato blight

Phylum Bacillariophyta (diatoms)

most diverse algal phylum over 10,000 marine and fresh water species, secrete silica tests (shells)

Phylum Chrysophyta (golden algae)

combination of yellow and brown pigments, unicellular species of ponds and lakes

Phylum Phaeophyta (brown algae)

multicellular marine algae usually known as sea weeds

*Macrocystis* is giant kelp along CA coast

**Kingdom Rhodophyta** (red algae)

mostly marine algae, mostly multicellular, possess unique red phytopigments

**Kingdom Chlorophyta** (green algae)

unicellular and filamentous algae, fresh water and marine

*Chlamydomonas* - flagellated unicellular fresh water alga

*Volvox* - a colonial flagellate, tendency toward multicellularity shown in the specialized reproductive cells

*Spirogyra* - a filamentous freshwater alga

**Kingdom Mycetozoa**

terrestrial organisms that feed by absorption of nutrients from dead organic matter

Phylum Myxogastriada (plasmodial slime molds)

Phylum Dictyostelida (cellular slime molds)

Phyla of undetermined affinities (pseudopodia present)

**Phylum Rhizopoda** (amoebas)

**Phylum Actinopoda** (radiolarians and heliozoans)

**Phylum Foraminifera** (foraminiferans)

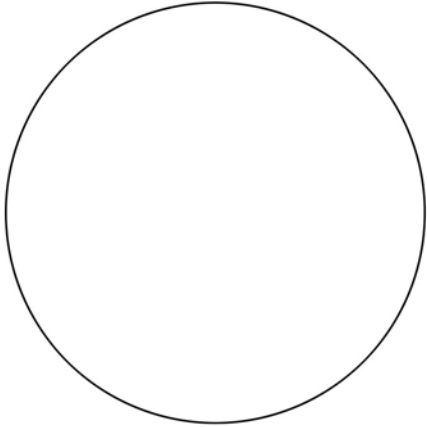
single-celled, calcium carbonate skeleton

**Biology 3B Laboratory: The Protista**

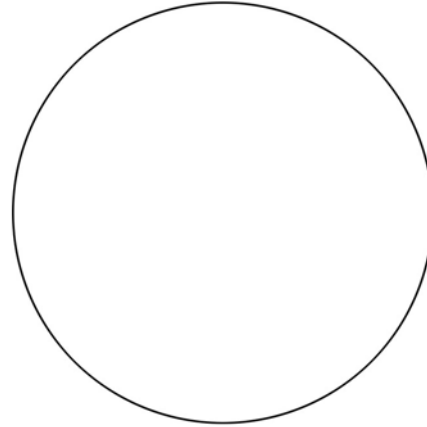
**Name:** \_\_\_\_\_

**Kingdom Euglenozoa**

Draw and label your *Euglena* here



Magnification: \_\_\_\_\_



Magnification: \_\_\_\_\_

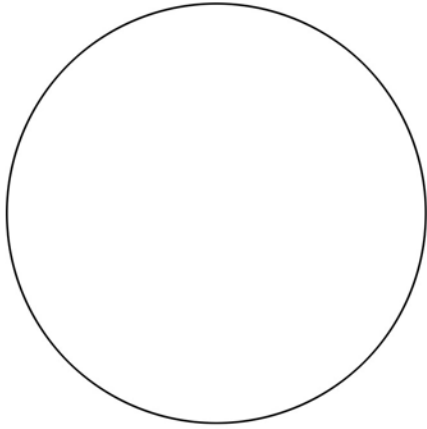
Record your *Euglena* lengths and calculate an average length in the following space:

In the space below draw the series of pictures that illustrate “euglenoid” locomotion

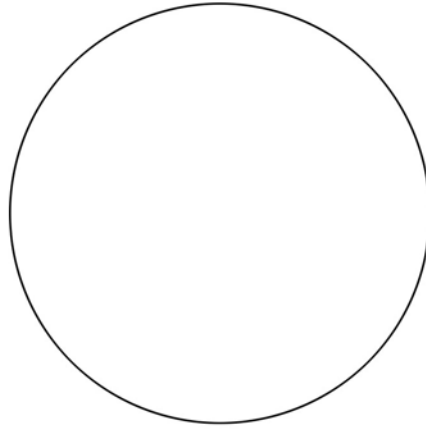
Write a description of the “euglenoid” locomotion:

**Kingdom Alveolata**

**Phylum Dinoflagellata**  
Draw and label Noctiluca and Ceratium

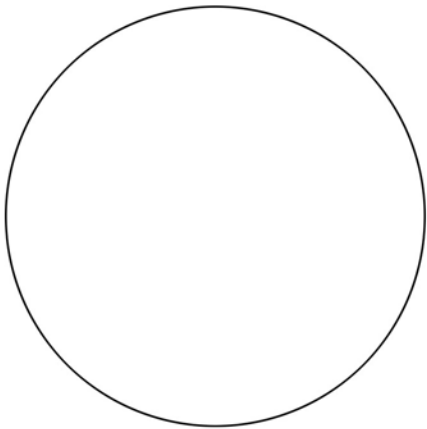


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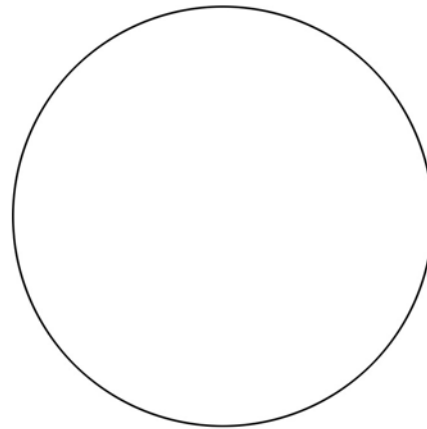


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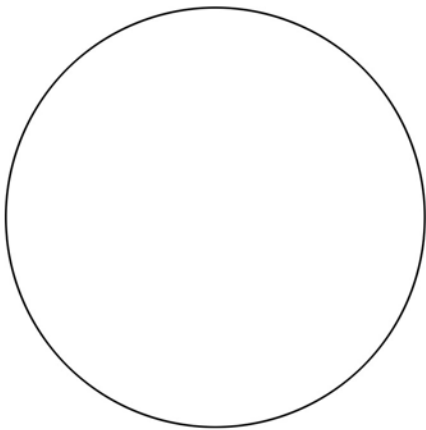
**Phylum Ciliophora**  
Draw and label Paramecium, Blepharisma and Stentor



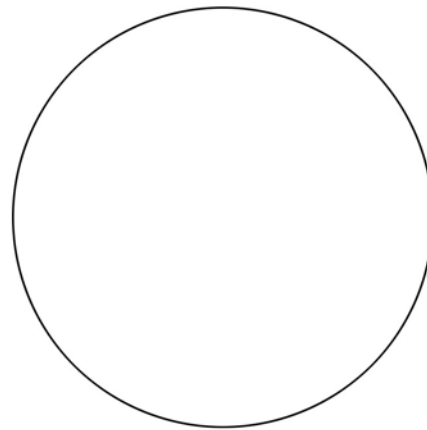
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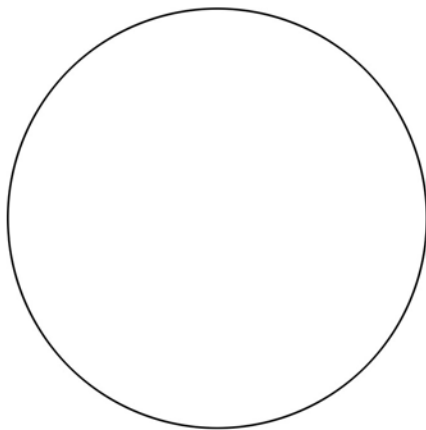
Describe the reaction of one of the ciliates to acid and salt  
Species used: \_\_\_\_\_

Acid

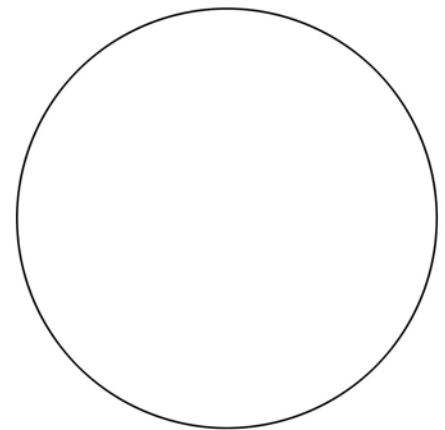
Salt

Phylum Stramenopila  
Kingdom Oomycota

Draw the potato blight organism infecting a potato leaf



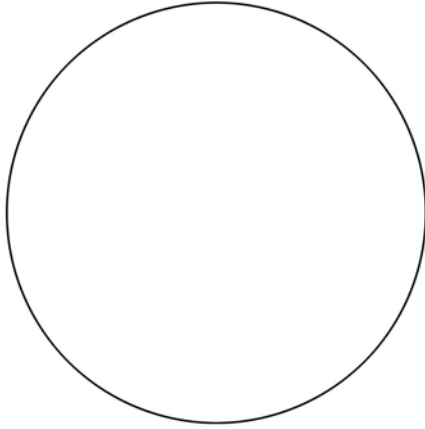
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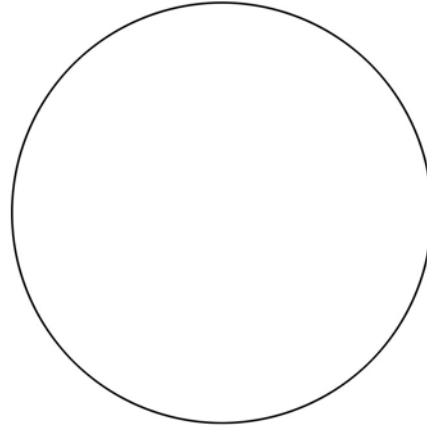
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Are there any examples of water mold infection in the plants around the Science/  
Math building?

**Phylum Bacillariophyta**  
Draw several diatoms here



Magnification: —



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**Phylum Phaeophyta**  
In the space below sketch some of the brown algae (be sure to jot down the species names) and complete a sketch of a typical brown algal life cycle.

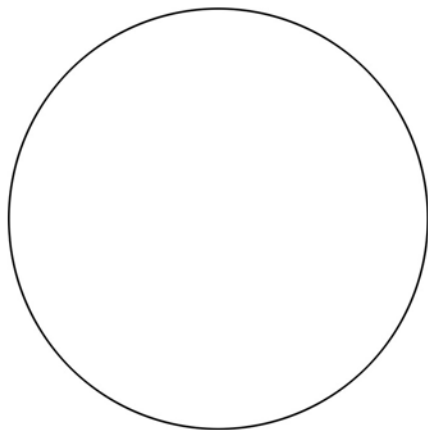
## Kingdom Rhodophyta

In the space below sketch a few of the red algae shapes.

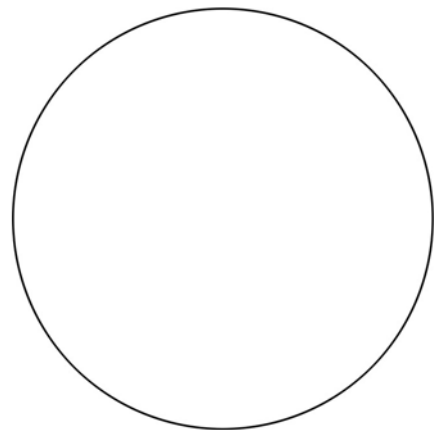
What is the role of the red algae in tropical coral reef systems?

## Kingdom Chlorophyta

Draw and label some of the *Volvox* colonies



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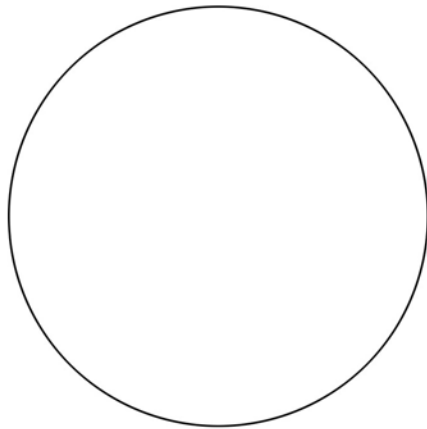
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In the space below draw one of the marine green algae species

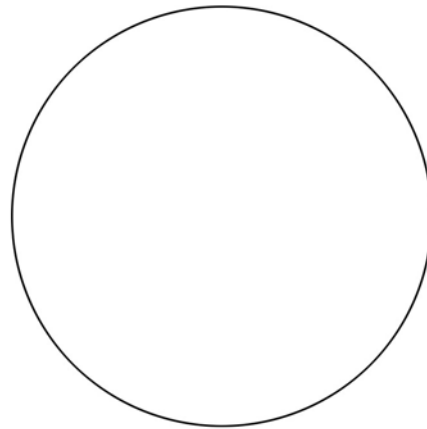
**Other Phyla**

**Phylum Rhizopoda**

Draw and label *Amoeba proteus*



Magnification: \_\_\_\_\_



Magnification: \_\_\_\_\_

Create six small sketches to illustrate *Amoeba* locomotion (thirty second intervals)

What is the speed of *Amoeba* movement (please show your work)

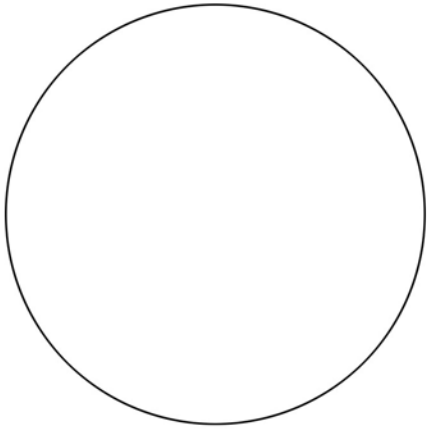
What is the "lifetime" of a single contractile vacuole? \_\_\_\_\_

What is the "lifetime" of a single contractile vacuole when the organism is suspended in distilled water? \_\_\_\_\_

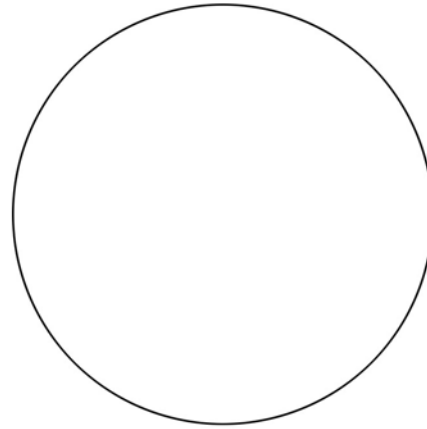
What can you conclude about the function of contractile vacuoles (regarding osmoregulation) in this organism?

**Phylum Actinopoda**

Draw radiolarian tests here



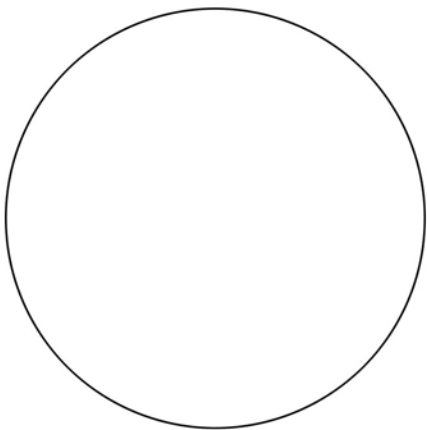
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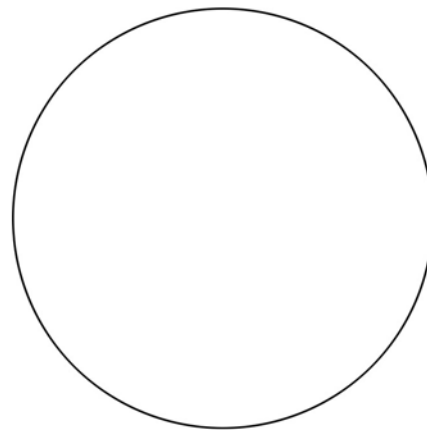
Magnification: —

**Phylum Foraminifera**

Draw some forams here



Magnification: —



Magnification: —

Complete the following table with information from lab, the text, or other research



Are there any characteristics in common among the phyla you observed?

The early protists are generally considered ancestral to the higher metazoan kingdoms (Plants, Animals, and Fungi). Which characteristics in which groups of protists suggest relationships shared with plants, animals, or fungi?

In your opinion do the protists represent a “cladistically” natural group (synapomorphies present)? Please discuss this in light of the observation made in this lab.