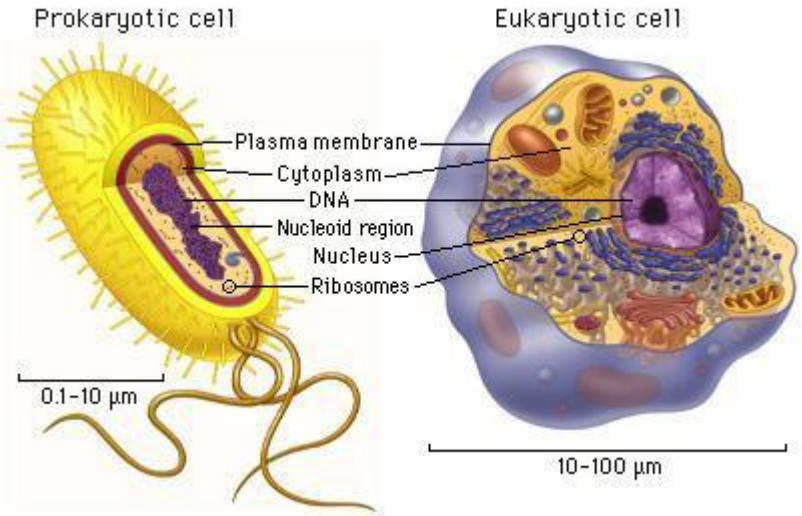
Protozoans and Algae Lab

Prokaryotic and Eukaryotic Cells



As you know, the building blocks of life are cells. **Prokaryotic** cells are those cells that do NOT have a nucleus. They mostly include bacteria and archaea. These cells do not have membrane-bound organelles. **Eukaryotic** cells are those that have a true nucleus. That would include plant, animal, algae, and fungal cells. As you can see, to the left, eukaryotic cells are typically larger than prokaryotic cells.

In today's lab, we will look at eukaryotic unicellular organisms, most of which are commonly found in pond water. When examining pond water under a microscope, the unpigmented, moving microbes will usually be protozoans. Greenish or golden-brown organisms will typically be algae.

Lab Objectives:

- Identify common species of protozoa
- Differentiate among the major classes of protozoa
- Identify common species of algae
- Examine different growth forms of algae

The Protists - Eukaryotic Organisms Kingdom Protista

Single-celled, eukaryotic organisms are called **protists**. They are grouped together in **Kingdom Protista**. Those protists that are animal-like are put in **Subkingdom Protozoa**, and the protists that are plantlike belong to **Subkingdom Algae**.

Protozoans are a large group of single-celled, usually microscopic, eukaryotic organisms, such as amoebas, ciliates, flagellates, and sporozoans. Protozoans (proto = first and zoa = animal) are

heterotrophic organisms that occupy marine, freshwater and terrestrial environments. Members of this group are generally characterized by their mode of locomotion; (1) **ameboid** – use pseudopods (Phylum Rhizopoda e.g. *Amoeba*), (2) **ciliate** – use cilia (Phylum Ciliophora e.g. *Paramecium*) and (3) **flagellate** – use flagella (Phylum Sarcomastigophora e.g. *Trypanosoma*). In addition, some protozoans also possess a **food vacuole** which is used to digest and absorb ingested materials and **contractile vacuoles** that function in expelling water. Reproduction in these organisms varies, but most genera reproduce either asexually or sexually.

Algae are unicellular or multicellular organisms (formerly classified as plants) that have chlorophyll but lack true stems, roots, and leaves. Algae include the seaweeds, diatoms, and spirogyra.

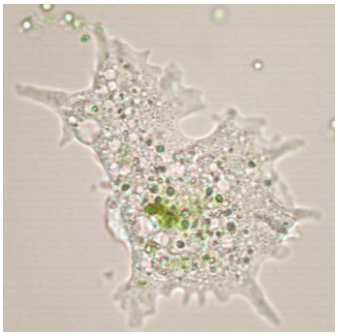
The Protists – Subkingdom Protozoa

Members of Subkingdom Protozoa, known as the **protozoans**, have a cell membrane (the pellicle) but lack cell walls. The nuclei are distinct, and the cells often have specialized organelles such as contractile vacuoles, mitochondria, flagella, and cilia. All protozoans produce **cysts** which are resistant dormant stages that enable the protozoans to withstand drought, heat, and freezing.

The protozoans we will look at today include the following:

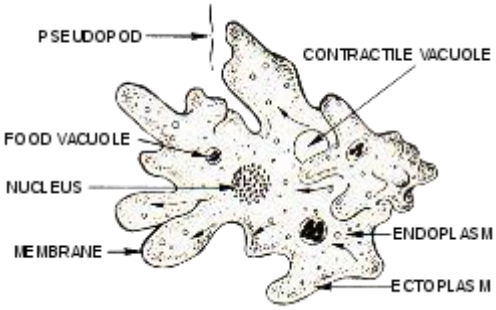
- Amoebas
- Paramecium
- Plasmodium
- Trypanosoma

Amoebas move by cytoplasmic streaming, having no motility structure. You will likely see some freshwater amoebas in the pond water, some of which may have tests or 'shells' that surround their cytoplasm. The flagellates have flagella or an undulating membrane for motility. The ciliates have cilia. The Apicomplexa have a unique arrangement of **microtubules**, called the apical complex (used in the takeover of the host cell), in the cell. This last class has most of the human and animal pathogens in it. The classes of protozoa are categorized by a variety of factors: cell architecture, motility structure, even hosts.



For our purposes, there are only 4 groups of protozoa that will be covered here: these groups are separated by motility and cell structure.

- Amoebas (representative: *Amoeba proteus*)
- Flagellates (representative: *Trypanosoma, Euglena*)
- Ciliates (representative: *Paramecium*)
- Apicomplexa (representative: *Plasmodium*)



Amoebas are unicellular **heterotrophs** (they must eat food, as do animals). They obtain food through **phagocytosis**, utilizing **pseudopods** to entrap their meals. Amoebas' most recognizable features include one or more nuclei and a simple **contractile vacuole** to maintain osmotic equilibrium. Like other eukaryotic organisms, they reproduce through asexually via mitosis and cytokinesis, not binary fission,

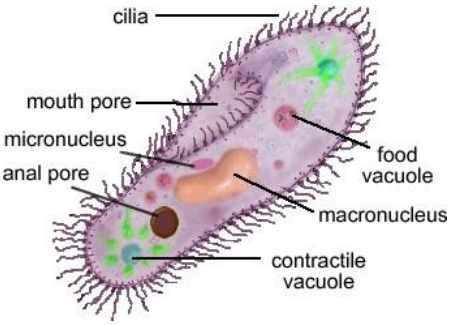
Watch this video of an amoeba in action!

http://youtu.be/W6rnhiMxtKU

Prepare a wet mount of the Amoeba and view under the microscope.

View a prepared Amoeba slide.

Paramecia are unicellular, ciliated **protozoans** (microscopic animals). Simple **cilia** cover the body, which allow the cell to move with a synchronous motion (like a caterpillar) at speeds of approximately 12 body lengths per second. There is also a deep oral groove containing inconspicuous tongue-like compound oral cilia used to draw food inside. They generally feed on bacteria and other small cells. Osmoregulation is carried out by a pair of contractile vacuoles, which actively expel water from the cell absorbed by osmosis from their surroundings. Paramecia are widespread in freshwater environments.



Watch this video of paramecium in action!

http://youtu.be/l9ymaSzcsdY

Prepare a wet mount of *Paramecium* and view under the microscope. Draw what you see.

View a prepared slide of *Paramecium*.

Observation of prepared slides of blood parasites:

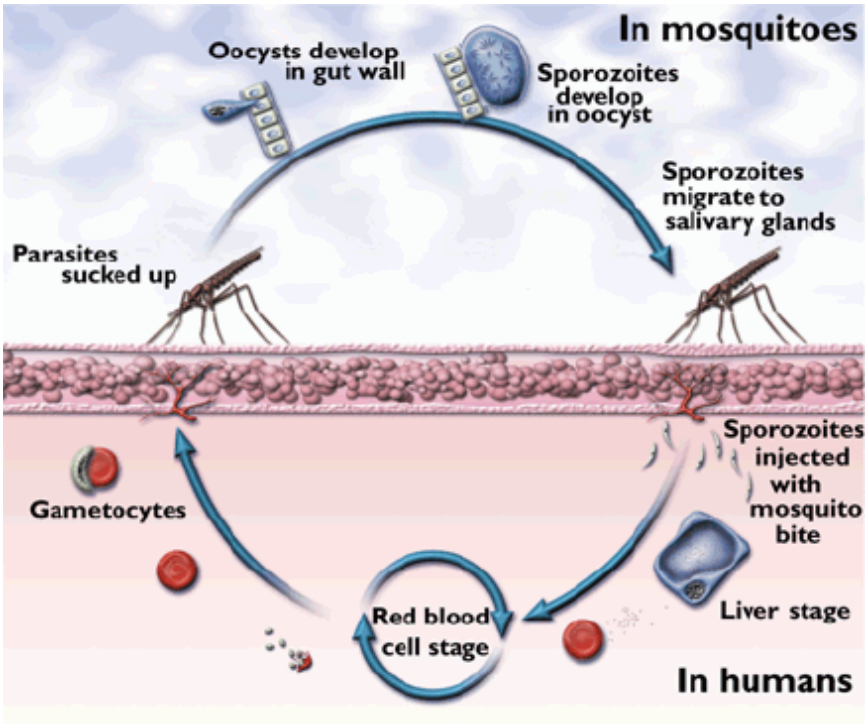
Trypanosoma *Plasmodium:*

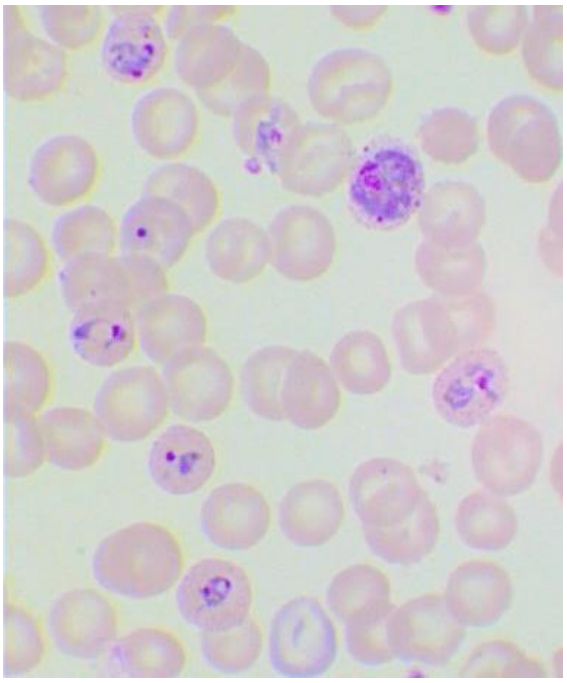
View a prepared slide of Trypanosoma.

Draw what you see:

Use bright field microscopy. Start with the 10X objective lens, ending up on 100X *Trypanosoma* will be easy to see: it is far larger than the red blood cells. However, *Plasmodium* will be difficult since the parasite will be inside of the RBCs. These are small protozoa, and so have to be viewed with 100X magnification.

Plasmodium is a genus of parasitic protists. Humans can become infected with at least 11 species of *Plasmodium*, resulting in **malaria.** Malaria translates literally as "bad air." We now know it to be caused by a parasitic protozoan transmitted to humans by *Anopheles* mosquitoes. The protozoan occupies human red blood corpuscles (RBC's), replicates inside them, and then destroys them as they escape to infect new RBC's. This lifecycle results in intermittent or remittent fever characterized by attacks of chills, fever, and sweating.



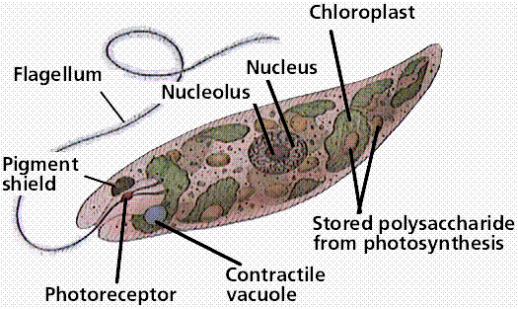


View a prepared slide of *Plamsmodium falciparum.*

The Protists – Subkingdom Algae

Subkingdom Algae includes all the photosynthetic, eukaryotic protists. Again, they lack the tissue differentiation found in plants. Algae may be unicellular, colonial, or filamentous. Algae have distinct, visible nuclei and chloroplasts. **Chloroplasts** are organelles that contain photosynthetic pigments which harvest the energy of sunlight to form carbohydrates through a process known as **photosynthesis**. There are 7 different divisions of algae, but we will examine only a selection of them:

- Euglenophyta (the euglenoids) – ex. *Euglena*
- Chlorophyta (the green algae) – ex. *Volvox, Ulothrix,* and *Spirogyra*
- Chrysophyta (the golden brown algae) – ex. Diatoms



Euglena are unicellular protozoans that almost always have **chloroplasts**. Although they photosynthesize (autotrophy), *Euglena* can also eat food by heterotrophy (like animals). They use a **flagellum** for locomotion.

Watch this video of euglena in action!

http://youtu.be/0rNI8Bos_BQ

View a prepared slide of *Euglena*. Draw what you see.

Phylum Chlorophyta: green algae

- Ancestors of Green Plants
  - ***Have cellulose cell walls***
  - ***Chloroplasts with Chlorophyll***
  - ***Store food as starch***
- Types of Algae
  - ***Chlamydomonas, single cells***
  - ***Spirogyra, filamentous***
  - ***Volvox, colonial***

Like vascular plants, green algae have chlorophylls a and b in addition to a variety of carotenes and xanthophylls that act as accessory pigments. Nutrition is autotrophic, with the reserve carbohydrates stored in plastids in the form of starch. Green algae exhibit a wide variety of thallus forms, ranging from single cells to filaments to parenchymatous thalli. In tropical and subtropical waters, many forms may be calcified.

Morphology

The green algae are well represented in the marine plankton and damp terrestrial environments, with many species occurring as unicellular organisms. These include motile vegetative cells that produce vegetative spores and/or gametes (e.g. *Chlamydomonas*), non-motile vegetative cells that produce flagellated reproductive structures (spores/gametes) (e.g. *Chlorococcum*), and lastly non-motile vegetative cells without motile reproductive structures (e.g. *Chlorella*).

Colonial green algae (e.g. *Volvox*) are not commonly encountered in the marine environment, though they are common residents of soil and standing freshwater. Colonies (synobia) are made up of distinct single cells held together by mucilage.

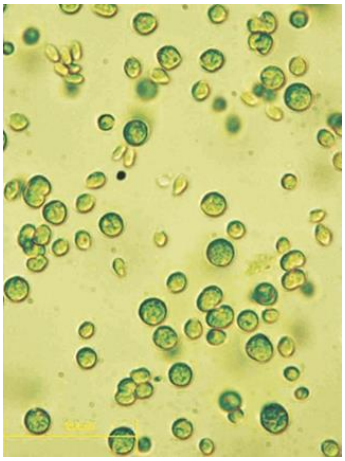
A giant evolutionary step was made in the green algae when the cells that resulted from a mitotic event remained contiguous. Connected cells enabled both multicellularity and the complex functions this required. Filamentous green algae are well represented in the sea either as unbranched, simple uniseriate (single thread) filaments of cells that show little or no thallus differentiation (e.g. *Chaetomorpha, Ulothrix*), or as branched filaments (e.g. *Cladophora*).

Unicellular Forms

Key Organism—*Chlamydomonas*, *Chlorella*

Unicellular forms can be motile or non-motile. *Chlamydomonas* is a single-celled motile alga commonly found in damp soil, ditches, and tide pools and occasionally in salt marshes. The organism is typically

egg-shaped and has a large cup-shaped chloroplast containing a proteinacious body—the pyrenoid, which functions in starch polymerization. The nucleus is often difficult to see because of the prominent chloroplast. *Chlorella* is a single-celled non-motile alga that often forms a symbiosis with aquatic invertebrates and protozoa. The cells contain a **stigma** (eyespot) that functions in the absorption of light. Reproduction in *Chlamydomona*s is usually asexual except during times of environmental stress, when the organism produces identically sized and shaped gametes (**isogamy**) for sexual reproduction.



Chlamydomonas

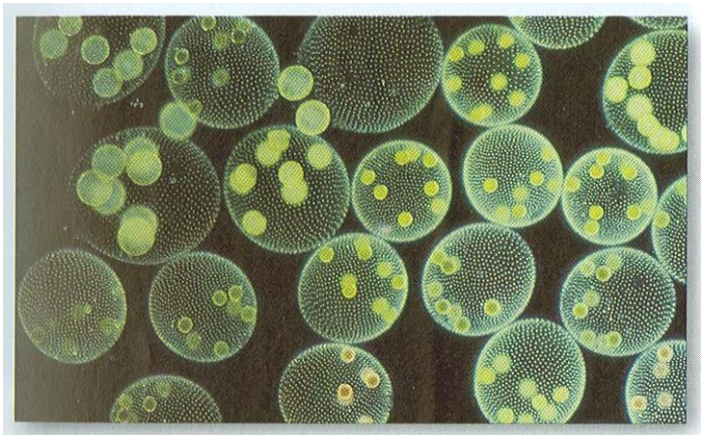
Examine the live and prepared slides of *Chlamydomonas* provided. **Draw** what you see.

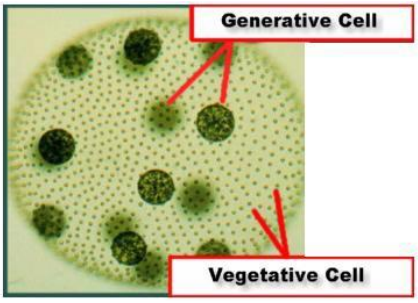
Colonial Forms

Volvox is a type of **green algae**. It forms **spherical colonies** of up to 50,000 cells. They live in a variety of freshwater habitats. Each mature *Volvox* colony is composed of numerous flagellate cells embedded in the surface of a hollow sphere. The cells swim in a coordinated fashion, with distinct anterior and posterior poles. The cells have eyespots which enable the colony to swim towards light. The spheres will break up with advanced age. The **generative cells** then grow into new colonies.

A *Volvox* colony can have up to 50,000 individual cells. The cells are arranged in a layer around the periphery of the colony. The two flagella of each cell are oriented out into the surrounding water. The inside of the colony is a

mixture of water and mucilage. New daughter colonies form within the colony. *Volvox* can reproduce asexually and sexually.



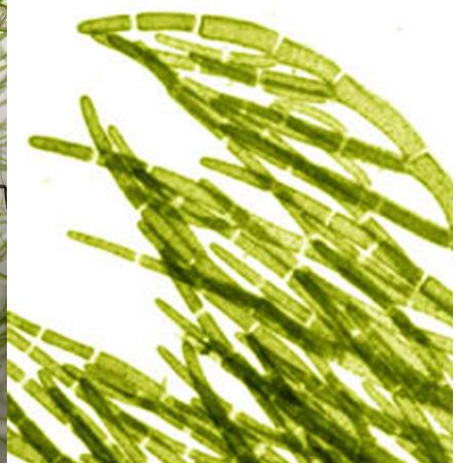


Watch this video of Volvox in action!

http://youtu.be/He9FSeGRi3A

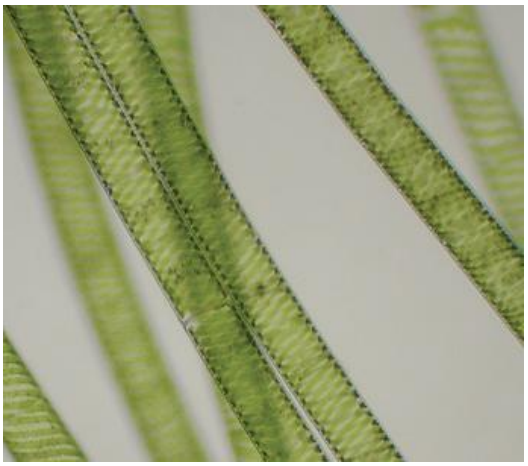
Examine the live and prepared samples of *Volvox* provided. **Draw** what you see.

Filamentous Forms



Cladophora is a **filamentous green algae**, generally found in fresh and marine water. *Cladophora* is found growing attached to rocks and other objects by means of a specialized cell, the holdfast. **Note** and **draw** the **holdfast** if it is present. **Draw** the extensive branching

Examine the live and prepared slides of *Cladophora* provided. **Draw** what you see.



Spirogyra is a **filamentous green algae** of the, named for the helical or **spiral arrangement ofthe chloroplasts**. It is commonly found in freshwater areas. *Spirogyra* measures approximately 10 to 100μm in width and may stretch centimeters long. This particular algal species, commonly found in polluted water, is often referred to as "pond scum". The cell wall has two layers: the outer wall is composed of **pectin that dissolves in water to make the filament slimy to touch** while the inner wall is of cellulose. The cytoplasm forms a thin lining between the cell wall and the large vacuole it surrounds. The chloroplasts are ribbon shaped, serrated or scalloped, and spirally arranged. In spring *Spirogyra* grows under water, but when there is enough sunlight and warmth they produce large amounts of oxygen, adhering as bubbles between the tangled filaments. The filamentous masses come to the surface and become visible as slimy green mats. This picture shows part of a gigantic single cell of *Spirogyra*. This organism is unusual because the chloroplasts form a long spiral inside the cylindrical cells.

Spirogyra is called a filamentous alga, because it grows as long, filament-like chains of cells. This body plan provides a ratio of surface area to volume, which is important because each cell must independently absorb nutrients and sunlight directly from the environment. Although the cells are linked together, they live more or less independently.

Examine the live and prepared slides of *Spirogyra* provided. **Draw** what you see.

Key points about green algae:

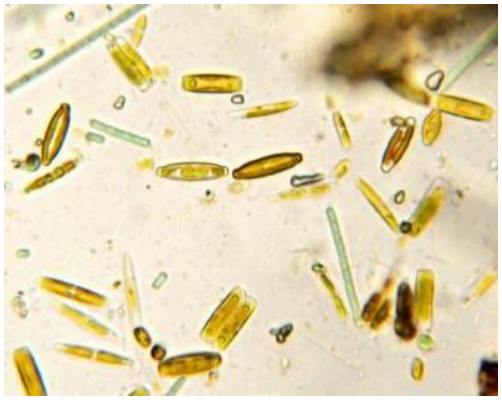
- Photosynthetic eukaryotes.
- Closely related to green plants.
- Many are unicellular, but some have simple multicellular body plans.
- Do not have differentiated tissues other than reproductive cells.

Because they lack differentiated tissues and other adaptations to life on land, they must live and reproduce in the water.

Chrysophyta: Diatoms

- Most numerous Algae in Oceans
- Cell wall contains silica dioxide, "silica shell"
- Similar Biochemically to Brown Algae above
- Economic Importance
- Abrasive surfaces, used in toothpaste, scouring powder etc.

Diatoms are a major group of **golden brown algae**, and are one of the most common types of **phytoplankton** (microscopic plants). Diatoms are **primary producers** within the food chain. Most diatoms are unicellular, although they can exist as colonies in the shape of filaments or ribbons, fans, zigzags, or stellate colonies. Diatom cells are characteristically encased within a unique cell wall made of **silica** (hydrated silicon dioxide) called a **frustule**. These frustules show a wide diversity in form, but **usually consist of two symmetrical sides with a split between them**, hence the group name.



Prepare a wet mount of diatomaceous earth and observe under the microscope. Draw what you see.

View a prepared Diatom slide.

Source:

http://programs.clarendoncollege.edu/programs/NatSci/Biology/Microbiology/micro%20online/Microbiology%20Protozoan%20lab.pdf