# Exercise 9 AN INTRODUCTION TO MICROSCOPIC ALGAE

### Introduction

Microscopic **algae** (singular **alga**) are classified here within the kingdom *Protista*, although some references divide them among other kingdoms including *Chromista* and *Alveolata*. Organisms categorized as algae are eukaryotic and most are **oxygenic photoautotrophs**, but some forms can also function as chemoheterotrophs. Algae that contain green **chlorophyll** pigments are important ecologically as producers of oxygen and because they serve as basal links in various food chains. Some algae exist as single cells, while others form colonies, coenocytes, or complex plant-like structures. Not all algae are microscopic, but we will focus primarily on those that are. The science or study of algae is called **Phycology**.

The classification of algae was initially based on a number of factors including; differences in pigments contained, food reserves present, structure of chloroplasts, type of flagellation, and the chemical composition of cell walls. More recently, studies involving the biochemical analyses of nucleic acids have provided new insights into algae **phylogeny** (evolutionary history), and suggest that the classification scheme most commonly used for these organisms should be changed. For this class, the microscopic algae will be organized into a number of groups (Phyla or Divisions) as follows:

# Chlorophyta:

The *Chlorophyta* or green algae are primarily fresh water forms, though some inhabit damp soil or similar habitats, and some are found in marine environments. Their cells are enclosed in walls of cellulose and pectin, they have single nuclei, and well organized chloroplasts. There are nearly 7000 species of green algae in 450 genera including *Spirogyra*, *Ulothrix*, *Oedogonium*, *Chlamydomonas*, *Cladophora* and multiple forms commonly known as **desmids**.

# Rhodophyta & Phaeophyta:

The *Rhodophyta* (red algae) and *Phaeophyta* (brown algae) are organisms often associated with marine habitats. (Brown algae are almost entirely restricted to the sea.) They are similar in structure to the green algae; but have, along with chlorophyll, pigments (carotinoids and/or phycobilins) that make them appear brown or reddish in color. Red algae are important to microbiology in that they are the source agar, the polysaccharide used to solidify most culture media.

#### Bacillariophyta:

The *Bacillariophyta* (commonly called **diatoms**) have complex cells consisting of two overlapping walls fitted together much like the halves of a Petri dish. These halves are called **frustules** and are composed largely of silica (glass). Diatoms are common in fresh and salt water and in damp soil. They are sometimes categorized in the phylum *Chrysophyta*, as golden or yellow-green algae. Several different genera of diatoms are represented on some diatom type slides.

# Dinoflagellata, Dinophyta or Pyrrophyta:

Organisms within the phylum *Dinoflagellata, Dinophyta* or *Pyrrophyta* are commonly known as **dinoflagellates**. Most are marine forms, but a few inhabit fresh waters. Many dinoflagellates are armored or covered with stiff cellulose plates called **thecae**, they have two flagella which beat within groves, one **transverse** (running around the cell) the other **longitudinal**. A number of dinoflagellates are **bioluminescent** (produce light), some can consume other cells, and some produce toxins that can cause paralytic shellfish poisoning or neurological symptoms in man. Dinoflagellates are often considered to be protozoa because some are strictly chemoheterotrophs and because the group is genetically similar to ciliates and apicomplexans. Examples include *Peridinium* and *Ceratium*.

# Haptophyta:

Organisms in the phylum *Haptophyta* (commonly called **coccolithophores**) are single-celled forms covered by calcium carbonate plates (or scales) called **coccoliths** that are generated internally. They are almost exclusively marine, form a large percentage of the earth's phytoplankton and contribute to the formation of chalk deposits. Recent increases in atmospheric carbon dioxide ( $CO_2$ ) levels have caused sharp increases in coccolithophore populations.

Note – Although *Euglena* and other green-colored, flagellated single-celled organisms commonly referred to as euglenoids are sometimes categorized within the phylum *Euglenophyta* (*Euglenophycophyta*); for this class, they will be categorized with the protozoa in the phylum *Euglenozoa*.

# Procedure

- A. Observe the prepared slides listed below. Note the shape and arrangement of cells, chloroplast shape when visible, and other distinctive features.
  - 1. Chlorophyta:
    - a. Chlamydomonas
    - b. Spirogyra
    - c. Ulothrix
    - d. Oedogonium
    - e. Cladophora
    - f. Desmids
  - 2. Bacillariophyta:
    - a. Diatom type slide

- 3. Dinoflagellata (Dinophyta or Pyrrophyta)
  - a. Peridinium
  - b. Ceratium
- 4. *Haptophyta*:
  - a. Observe scanning electron micrographs of coccolithophores
- B. Make wet mounts of and observe the examples of living algae provided. Notice variations in cell structure and arrangement, chloroplast shape, and other features that might be used for classification.
  - 1. Chlorophyta:
    - a. Chlorella
    - b. Cladophora
    - b. Ulothrix
    - c. Volvox
    - d. Hydrodictyon
    - e. Haematococcus
  - 2. Dinoflagellata:
    - a. Peridinium

**Note** - Living cultures may contain organisms other than algae. Consider how you can distinguish algae from other organisms (such as cyanobacteria) that may be present. You may wish to refer to the booklet "<u>How to Know the fresh water Algae</u>" in order to identify some of the organisms present.

C. Obtain a small sample of pond scum or wet soil and prepare a wet mount. Look for living diatoms and note their shape, coloration and motion.

# Questions

- 1. Which of the living algae you observed demonstrated motility? What was their mechanism of locomotion?
- 2. Which of the algae are unicellular in form? Which are filamentous?
- 3. Which of these algae contain green chlorophyll pigments?



Figure 9.1 – Some representative examples of filamentous fresh-water algae.



Figure 9.2 – Some representative examples of common unicellular algae.

# NOTES, OBSERVATIONS & ADDITIONAL INFORMATION

# Exercise 10 AN INTRODUCTION TO PROTOZOA

#### Introduction

**Protozoa** (singular protozoan) are classified here within the Kingdom *Protista*, although some references divide them among multiple other kingdoms e.g., *Alveolata, Crecozoa, Radiolaria, Diplomonadida, Parabasala* etc. Protozoa can best be described as single-celled, animal-like organisms. They are eucaryotic and often have complex structure with a variety of specialized organelles. Protozoa are generally considered to be **chemoheterotrophs**, but some genera such as *Euglena* can function as **photoautotrophs**. Some protozoa function as predators, some are saprotrophs (use dead or decaying material for food), and some are parasites that live within and obtain nutrients from other living organisms. Almost all protozoa are motile, and their classification was initially based primarily on their mode of locomotion. More recently, studies involving the biochemical analyses of nucleic acids have provided new insights into protozoan phylogeny, and suggest that the classification schemes most commonly used for these organisms do not accurately represent phylogenetic relationships. For this class, in the interest of simplicity, the protozoa will remain within the kingdom *Protista* and be organized into a number of phyla as follows:

#### Phylum Archaezoa or Diplomonadida (Diplomonads):

Organisms categorized as *Diplomonadida* were formerly considered anchient forms (*Archaezoa*) because they lack mitochondria, golgi bodies and peroxisomes and were thought to have evolved before the **endosymbiotic** event giving rise to mitochondria took place. Current taxonomists believe these organisms are descendants of typical eukaryotes that have lost their organelles during more recent times. Many Diplomonads live within the digestive tracts of other organisms and some are parasites, e.g., *Giardia*. They typically have two equal sized nuclei and multiple flagella.

#### Phylum Archaezoa or Parabasala (Parabasalids):

Organisms categorized as *Parabasala* also lack mitochondria and were formerly grouped with the diplomonads as anchient (*Archaezoa*). These organisms have only one nucleus, but each one also has a **parabasal body**, a structure similar to the golgi. Parabasalids often live symbiotically within other organisms, but are not parasitic. Examples include *Trichonympha*, heavily flagellated forms found within the guts of termites, and *Trichomonas*, organisms found living in the human vagina.

#### Phylum Amoebozoa (Amoebae):

The *Amoebazoa* are organisms that move by means of protoplasmic extensions called **pseudopodia** or false feet. They obtain food through **phagocytosis**, and often consume other eukaryotic organisms, e.g., diatoms, desmids, etc. Because their pseudopodia can extend in various directions, amoebae appear in a variety of shapes. Although most live freely in fresh and salt water, some are parasitic and some cause serious disease symptoms in humans. Example amoebae include *Amoeba proteus*, *Entamoeba histolytica*, *Acanthamoeba* and the slime molds described earlier with fungi.

## Phylum Amoebozoa or Radiolaria (Radiolarians):

The *Radiolaria* are amoeba-like organisms with thin, thread-like pseudopodia supported by stiff microtubule bundles. They form ornate external coverings called **skeletons** from glass (silica dioxide - SiO<sub>2</sub>) and extend their pseudopodia outward through the multiple openings like spokes in spherical wheels. Radiolarian skeletons provide protection and give the many different genera a distinctive appearance. Collections of these skeletons form a sedimentary rock type called radiolarian chert. Many different types are visible in a **radiolarian strew**, a concentration of organisms collected from water.

## Phylum Apicomplexa (Apicomplexans):

The *Apicomplexa* are obligate intracellular parasites (hypotrophs) that are not motile in their mature forms. All are animal parasites and several are important human pathogens. The group is characterized by the presence of specialized organelles forming a complex at the apical end of infective stage cells (hence the name apicomplexa). These organelles contain enzymes that help the protozoa penetrate host tissues. Important example apicomplexans include *Plasmodium* and *Toxoplasma*. These organisms are sometimes referred to as sporozoans (Class *Sporozoea*).

### Phylum Ciliophora (Ciliates):

Organisms within the phylum *Ciliophora* form a diverse group with many morphologically distinctive genera. All are equipped with **cilia**, but some have these arranged in rows covering their entire surface, while some have them only in patches, and others have them arranged in tufts called **cirri**. Although cilia are used primarily for locomotion, they are also used to sweep food toward and into the **cytostome** or cell mouth, located within an oral funnel or groove in some genera. All ciliates have two nuclei, one large (macronucleus) and one small (micronucleus). Most ciliates live freely in water environments where they feed on bacteria and small eukaryotic organisms, but some are parasites. Examples include *Paramecium*, *Vorticella*, *Stentor* and *Balantidium*.

#### Phylum Euglenozoa

Organisms classified within the phylum *Euglenozoa* include the **euglenoids**, formerly categorized as algae because they are primarily photoautotrophic, and the **hemoflagellates** or blood parasites. These two groups are not morphologically similar, but recent findings show that all share common ribosomal-RNA nucleotide sequences, and have the same disc-shaped mitochondria. The Euglenoids typically contain green chlorophyll pigments but also have red-colored pigments (an eye-spot) at their anterior end. Many can change shape, and all move by means of one anterior flagellum that pulls them through their environment. Although most euglenoids are photoauto-trophs, they can live as chemoheterotrophs if light is not available. The hemoflagellates have a single flagellum attached to their cell surface by an undulating membrane (i.e., a double layer of cell membrane) and are transmitted from one host to another by biting insects. Like the euglenoids, hemoflagellates typically have a pigment spot at their anterior end. Example organisms include multiple free-living forms in the genus *Euglena* and parasites in the genus *Trypanosoma*.

**Note** – The Phyla *Ciliophora, Apicomplexa* and *Dinoflagellata* are sometimes categorized together within the *Alveolata*, a taxonomic rank variously listed as a kingdom, a sub-kingdom or a super-phylum depending on the reference.

## Procedure

1. Observe the prepared slides of Protozoa as indicated below: Note the cell shape and structures characteristic to each group.

### Phylum Archaezoa or Diplomonadida

- a. Giardia lamblia (intestinalis) trophozoites
- b. Giardia lamblia (intestinalis) cysts

## Phylum Archezoa or Parabasala (Parabasalae)

a. Trichomonas vaginalis

### Phylum Amoebozoa

a. Amoeba proteus

### Phylum Amoebozoa or Radiolaria

a. Radiolarian type slide

# Phylum Apicomplexa - Class Sporozoea

a. Plasmodium vivax (inside RBCs)

### Phylum Ciliophora

a. Paramecium caudatum, aurelia, or bursaria

# Phylum *Euglenozoa*

- a. Euglena gracilis
- b. Trypanosoma lewisi or gambiense
- 2. Prepare wet mounts of the various protozoa samples provided, observe them under the light microscope and try to identify them as to phylum.
- 3. Make wet mounts of some of the natural infusions provided and try to identify the protozoa present as to phylum.

# Questions

- 1. Most protozoa are nutritionally categorized as \_\_\_\_\_\_ but some forms are capable of using light energy and inorganic carbon. Which protozoa often contain chloroplasts and function as photoautotrophs?
- 2. Protozoa are single-celled organisms with eukaryotic cells. What types of organelles are readily visible within or on these organisms?
- 3. In which living forms are contractile vacuoles evident? What is their function?
- 4. Organism called *Trichonympha* are catagorized within which protozoan phylum? Where are these organisms found?
- 5. Which protozoa have more than one nucleus?



Figure 10.1 – Illustrations of Some Representative Protozoa



