Chapter 9

Overview of Autotrophic and Heterotrophic Protists

Objectives

Protista. Establish familiarity with the Protista. Understand some aspects of the importance of protists. Know the primary differences between autotrophs and heterotrophs. Define plankton and describe how they are important.

Autotrophic Protists. Know the taxa of autotrophic protists and the general characteristics used to determine these groups. Understand endosymbiosis. Understand the similarities and differences between zygotic meiosis and alternation of generations (a.k.a. sporic meiosis). Know the general morphological gamete forms and general types of sexual reproduction of protists. Know the similarities of and differences between green algae and plants. Become familiar with the diversity of green algae, and their structures, and life cycles. Understand the differences between and distinguishing characteristics of Chlorophyta (green algae), Rhodophyta (red algae), and Phaeophyta (brown algae, a photosynthetic heterokont).

Introduction to Protists

Protista comprises an assortment of primitive unicellular, colonial, and multicellular eukaryotes including simple photoautotrophic¹ organisms (*i.e.*. algae), protozoa (mobile, heterotrophic, and animal-like, *e.g.* Amoeba), and simple heterotrophic² organisms (*e.g.* slime molds and Oomycetes). This course will primarily focus on photoautotrophic protists. Three taxa of multicellular organisms, Plantae, Animalia, and Fungi, evolved from protists although protists do not have the distinguishing characteristics of any of the other kingdoms.

Protists are important in ecology and in studies of evolution. Photosynthetic protists in aquatic environments have similar importance as plants in terrestrial environments. Most protists are aquatic and are a major food source in aquatic habitats. Humans, in many cultures, eat protists, especially algae³. In addition, protists can cause disease and ecosystem disturbance. For example, when their habitat is disturbed, dinoflagellate populations explode, resulting in a "bloom" or in "red tide," that results in large quantities of toxic compounds accumulating and causing further disturbance of the ecosystem⁴.

Protists are diverse and polyphyletic. They vary in size from one cell to tens of meters long (*e.g.* kelp, brown algae (Phaeophyta)). When present, motility of protists may be effected by flagella, cilia, or amoeboid movement. Protists may or may not have cell walls. The asexual and sexual reproductive cycles of protists are varied and include all three types of general life cycles; however, in general, protists lack complex reproductive structures. Three major types of gamete morpohologies in sexual reproduction⁵ are observed in protists: isogamy (gametes are of equal size, shape, and motility), oogamy (one gamete is usually larger and is always nonmotile), and

¹ photosynthetic and therefore do not require an external source of reduced carbon

 $^{^{2}}$ depend on external sources of reduced carbon.

³ p. 300

⁴ p. 302

⁵ Fig. 15-15

anisogamy (dissimilar gametes that do not meet the definition of oogamy). Anisogamy literally means "not the same" but it doesn't mean they both are flagellated, for example, although they are often portrayed this way.

Autotrophic Protists

Photosynthetic protists were classified historically on the basis of pigmentation, cell-wall composition, form and location of carbohydrate storage and other features. Although our understanding has been vastly improved by molecular analysis, the general groups are still valid. Four of the major taxa are Chlorophyta (green algae), Rhodophyta (red algae), Phaeophyta (brown algae), and Chrysophyta (diatoms). The word alga is not a formal taxonomic term and is often used to include cyanobacteria (or blue-green algae) even though cyanobacteria are prokaryotes. Cyanobacteria are the evolutionary progenitors of all plastids, through a single endosymbiotic event. Photosynthetic protists are extraordinarily diverse in sexual reproduction and all three of the general sexual reproduction methods, as well as complex variations, are presented.

Green Algae (Chlorophyta). Chlorophyta are a diverse group. Plants evolved from a green alga. In general, green algae and plants contain chlorophylls *a* and *b*, store starch in plastids that have grana and two limiting membranes, and have cell walls made of cellulose; however green algae, in general, do not have complex reproductive structures as in plants and none are embryophytes. Green algae may be unicellular (*e.g. Chlamydomonas*), colonial (*e.g. Volvox*), filamentous (*e.g. Spirogyra*), "semi-filamentous" (*e.g.* desmids), or multicellular (*e.g. Chara* and *Nitella*). Green algae undergo asexual and sexual reproduction.

Specimen 1: Chlamydomonas

- 1. Create a slightly raised coverslip by doing the following. Wipe a thin layer of petroleum jelly on the back of your hand or on a paper towel. Gently, scrape two opposing edges of a coverslip through the thin layer of petroleum jelly.
- 2. Lower the coverslip, petroleum side down, onto a slide to create a small gap between the coverslip and slide.
- 3. Place a drop of *Chlamydomonas* wild-type culture under the coverslip.
- 4. Observe under 100X or 400X and record observations below.
- 5. Repeat with the *Chlamydomonas* mutant and record your observations below.
- 6. Know compare what you saw with the wild-type and the mutant. How do you think the mutation has affected the *Chlamydomonas*?
- 7. Add a drop of methyl cellulose to the wild-type culture, mix well, and add a coverslip. Methyl cellulose slows the movement of *Chlamydomonas*.

- 8. Observe under 400X. Observe the single large chloroplast in each cell.
- 9. Try to observe the orange "eye-spot" (stigma) that is involved in light sensing.
- Dim the microscope light to observe the pair of anterior flagella that provide motility. Although flagella (~0.3µm) are below the resolution (~1µm) of the microscopes used in this course, they are visible by optical effects.

Chlamydomonas reproduces asexually and sexually⁶. In our example, sexual reproduction is through zygotic meiosis; thus, except for the zygote, it is haploid. All cells—normal vegetative cells and cells that are gametes—are similar; therefore, it is is isogamous. Asexual reproduction by cell division occurs most frequently and sexual reproduction is induced by unfavorable conditions. When cells are exposed to unfavorable conditions, such as nitrogen deprivation, (+) or (-) haploid strains of cells produce, through mitosis, (+) or (-) haploid gametes respectively. Gametes then pair anteriorly, undergo plasmogamy, then karyogamy, resulting in a diploid zygote, the zygospore. The zygospore has a thick, protective wall that can withstand unfavorable conditions. When favorable conditions return, the diploid nucleus of the zygospore undergoes meiosis (zygotic meiosis) forming four haploid cells.

Specimen 2: Volvox, a colonial green alga

 $Volvox^7$ is the dead-end evolutionary pinnacle of a colonial green alga that is based on *Chlamydomonas*-derived cells. The number of cells in the colony is specific to the particular species and ranges from 500-60,000. The cells are connected by cytoplasmic connections that allow cell-to-cell communication. The colonies are hollow and the cells on the periphery are biflagellate. The colony moves by synchronized beating of the flagella. Each cell has an eye-spot that detects light. Some cells are specialized for sexual reproduction through oogamous zygotic meiosis. Some cells are specialized for asexual reproduction and, thus, undergo mitosis to form small daughter colonies within the parent colony. When daughter colonies are mature, they breaks from the parent colony, increase in size by cell growth, and become free-living.

- 1. Prepare a raised coverslip as for the previous specimen.
- 2. Place a drop of *Volvox* culture under the coverslip.
- 3. Observe the spherical mobile colonies at 100X and record observations below. Look for colonies at different stages of the life cycle. Do you see daughter colonies? Do you see movement?
- 4. Under dim light, try to observe cytoplasmic connections, flagella, and the eye-spots of the cells. (Although, these may be below the resolution of the microscopes). You may want to add methyl cellulose to make closer observation easier.
- 5. Draw a colony of *Volvox* at 100X or 400X and label cells, daughter colonies, and flagella.

⁶ Fig. 15-41

⁷ Fig. 15-42

Specimen 3: *Spirogyra*, a filamentous green alga

- 1. Place a few filaments of *Spirogyra* on a slide and make a wet mount.
- 2. Observe under 100X. Observe the row of cells that comprises each filament.
- 3. Observe the spiral-shaped chloroplast(s) within each cell.
- 4. On each chloroplast, observe the pyrenoids, which were present, but not visible, in *Chlamydomonas* and *Volvox* chloroplasts also. The major functions of pyrenoids are to synthesize and store starch.
- 5. Observe the nucleus in each cell.
- 6. Draw and label the cells, cell walls, chloroplasts, pyrenoids, and nuclei of a filament of *Spirogyra* at 400X.

Specimen 4: Sexual reproduction of Spirogyra

*Spirogyra*⁸ undergoes asexual reproduction by cell division and fragmentation and also undergoes sexual reproduction by zygotic meiosis. During sexual reproduction, a conjugation tube forms between two filaments. Fertilization occurs either in the tube or after a gamete migrates through the tube into the other filament. The zygote becomes surrounded by a thick wall that enables it to survive in harsh conditions.

- 1. Observe a prepared slide of *Spirogyra* undergoing sexual reproduction under 100X. Notice that the slide shows the progression through sexual reproduction.
- 2. Note the pair of filaments arranged side-to-side. Each filament is composed of haploid cells. One filament is of the (+) type and the other is of the (-) type.
- 3. Observe the conjugation tube that forms between two filaments. Following fusion, the cytoplasm of the cells combine allowing the genetic material to fuse to form a diploid zygote that will, when conditions are favorable, undergo meiosis.

⁸ Fig. 15-52

4. Draw at least *two* stages of conjugation. Label gametes, conjugation tube, and zygote. Indicate haploid and diploid parts.

Earlier stage in sexual reproduction (_____X)

Later stage (_____X)

Specimen 5: Desmids, semi-filamentous green algae

Desmids⁹ are found in abundance in peat bogs. Desmids have two sections or semi-cells that are joined by a narrow isthmus. Cell division and sexual reproduction are similar to the related *Spirogyra*.

- 1. Place a drop of the mixture of desmids on a slide and add a coverslip.
- 2. Observe under 100X. Several genera of desmids are represented.
- 3. Observe the two sections of each cell. Each semi-cell, or half-cell, contains a chloroplast and pyrenoid, but they share a single central nucleus.
- 4. Draw and label the half-cells, chloroplasts, and nucleus (or indicate where the nucleus would be found) of two different desmids at 400X.

Specimen 6: Complex multicellular green algae, *Chara*¹⁰ and/or *Nitella*

As evolutionary precursors to plants, complex, multicellular green algae have features that are similar to plants. Some similarities, in addition to the similarities for all green algae, between complex, multicellular green algae and plants include presence of complex reproductive organs, node-like structures, and apical growth. However, the complex green algae are not embryophytes, a distinguishing characteristic of all plants.

- 1. Cut a small portion of the *Chara* "stem" and place on a drop of water on a slide. Notice that the body is brittle. This is caused by CaCO₃ (calcium carbonate) encrustation in the cell walls.
- 2. Observe the nodes, internodes, and branches.
- 3. Observe under the dissecting microscope at around 10X-20X.
- 4. Try to find and focus on the slightly red structures, which are complex reproductive structures termed gametangia, on the main filament or the branches. The gametangia make sperm or eggs, by mitosis, that are released into the surrounding water. External fertilization is followed by the formation of a diploid zygote that then undergoes meiosis.
- 5. Draw and label the main filament, branches, nodes, and gametangia of Chara.

Preserved specimens of other interesting green algae

Observe the preserved specimens of *Acetabularia* (mermaid's wine glass) and *Valonia* ("sea grass"). These are marine green algae and are coenocytic, meaning it is multinucleate. Notice how large a single cell can be!

Observe the preserved specimen of *Ulva lactuca* (sea lettuce). This marine green alga undergoes alternation of generation.

Red Algae (Rhodophyta)¹¹. Unlike green algae, few red algae are unicellular. The chloroplasts of red algae closely resemble cyanobacteria. Chloroplasts of red algae contain abundant linear light-harvesting pigments, which mask chlorophyll a (red algae do not have chlorophyll b). These linear pigments give red algae their distinctive color and are well-suited for

¹⁰ Fig. 15-56

¹¹ Figs. 15-29; 15-30

absorption of the wavelengths of light that penetrate deep waters where red algae are found in abundance. Red algae reproduce asexually by discharging spores and sexually by what can be exceedingly complex multiphase means.

Preserved specimens of red algae

Observe the coralline red algae, which are predominately.

Observe the dried edible sea-weeds, which are important ingredients of oriental cuisine.

Brown Algae (Phaeophyta)¹². Brown algae are heterokonts, which have two unequal flagella at some stage in the life cycle. Brown algae undergo either alternation of generations¹³ or gametic meiosis¹⁴. Brown algae range in size from microscopic to the largest of all sea-weeds, such as kelp and *Sargassum*; however, none are unicellular or colonial; all are complex. Brown algae have chlorophylls a, but not chlorophyll b. The light-harvesting pigment fucoxanthin gives brown algae their distinctive brown color.

Preserved specimens of brown algae

Observe the specimens of Fucus. Notice the blades and floats (air bladders), which keep the alga on the surface of the water.

Planktonic Protists

Planktonic organisms inhabit the water column of bodies of water and move with the currents.

Dinoflagellates¹⁵. Most dinoflagellates are unicellular biflagellates that live in marine and fresh-water habitats. Dinoflagellates have a characteristic complex, armor-like cell covering. Most dinoflagellates are autotrophic, others are heterotrophic or osmotrophic. Dinoflagellates undergo sexual and asexual reproduction.

Diatoms¹⁶. Diatoms are unicellular, colonial, or filamentous autotrophic organisms that live in marine and freshwater habitats. Diatoms are heterokonts, but typically lack flagella, except on gametes. Diatoms have characteristic walls made up of polymerized, opaline silica and consist of two overlapping halves.

Specimen 7: Planktonic Algae, Dinoflagellates and Diatoms

1. Observe a water sample under 100X and 400X.

¹² Figs. 15-23; 15-24 ¹³ Fig. 15-27

¹⁴ Fig. 15-28

¹⁵ Fig. 15-5

¹⁶ Fig. 15-20

2. Draw a few dinoflagellates and diatoms (at least two of each) that are present in the sample.

Dinoflagellates

Diatoms

Observations of dinoflagellates movement, color, symmetry, diversity, etc.

Heterotrophic Protists¹⁷

Oomycetes and Myxomycetes are examples of heterotrophic protists that may be free-living or parasitic. Superficially, these protists resemble fungi; however fungi have several distinctive characteristics. Aquatic and terrestrial forms of Oomycetes and Myxomycetes exist. The life cycle of Myxomycetes¹⁸ is interesting because it has an animal-like amoeboid or plasmodial phase (no cell wall) and a fungus-like phase that produces spores. The plasmodial phase may grow into massive structures that span hundreds of square mile.

Heterotrophic Eukaryotes next week

Specimen set-up today for next week: Spore printing

- 1. Obtain pieces of black and white paper, each about the size of your mushroom cap, and in the corner of each, write your full name.
- 2. Arrange the paper and mushroom so that half of your mushroom cap is on the white paper and half is on the black paper. Cover your mushroom with a piece of plastic (e.g. a beaker) to prevent dehydration. Mushrooms are basidiomycete fungi. The underside of the cap has gills or pores from which are released spores. Read the lesson for next week for more information. You will be looking more closely at the spore-producing structures next week.

¹⁷ pp. 309-312; 340-343 ¹⁸ Fig. 15-58

Specimen set-up today for next week: *Rhizopus* culture

- 1. Obtain a plastic bag.
- 2. Place your piece of bread into the plastic bag.
- 3. Close the bag ~80% (instead of closing completely, 100%).

Questions

- 1. How are Chlorophyta similar to plants? How do Chlorophyta differ from plants?
- 2. Describe two ways that the life cycle of *Chlamydomonas* is different from the life cycle of plants.
- 3. Chlorophyta, Phaeophyta, and Rhodophyta all have chlorophyll *a*, what does that tell you about the photosynthesis in these organisms?
- 4. Describe endosymbiosis in the context of the theory that chloroplasts originated from cyanobacteria.