Algae

Members of the Kingdom Plantae are not the only organisms that are photosynthetic. Photosynthetic organisms not in the Plant Kingdom, traditionally referred to as “algae” are typically aquatic and members of the traditionally recognized eukaryotic Kingdom Protista (where all simple eukaryotic organisms go) or the Domain Bacteria. The phylogenetic tree below provides an overview of the major lineages of life and how they are thought to be related to one another: the 7 groups numbered below are the 7 major lineages of photosynthetic organisms and they’re obviously not all that closely related to one another.

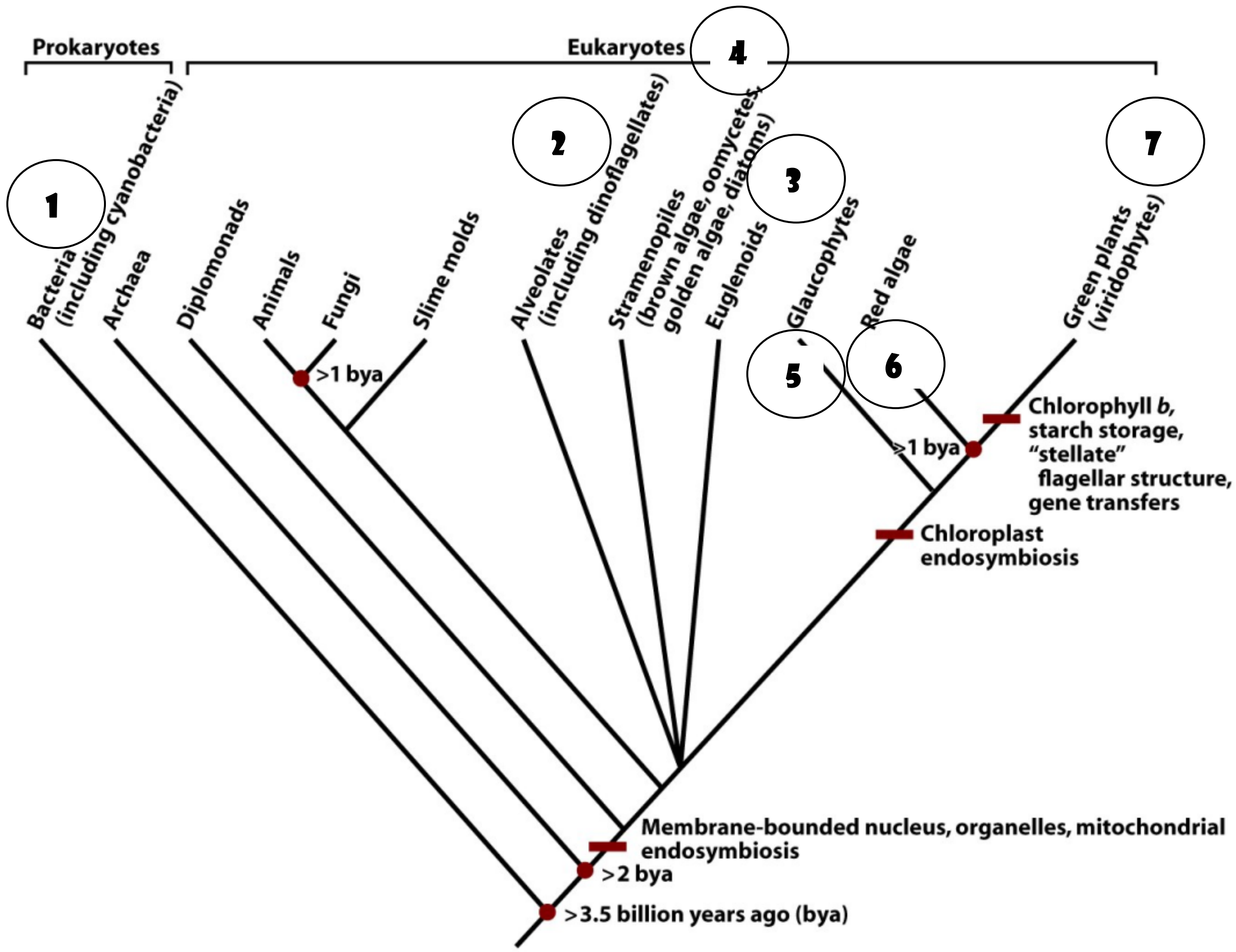

Figure 15-3
Biology of Plants, Seventh Edition

Adapted from Fig. 15-3 in Raven et al. (2007). A phylogenetic tree based on DNA sequences, showing 7 major groups of photosynthetic organisms.

"Algae" (or singular "alga") is historically a term applied to an unnatural assemblage of photosynthetic organisms that are usually morphologically simpler than members of the plant kingdom (the plant kingdom including bryotphytes, pteridophytes, and seed plants). Some algae are unicellular, but others are larger and multicellular organisms that are called "seaweeds". Within the algae you can see the major trend in the evolutionary development of multicellularity reflected in the diversity of organismal forms: i.e., unicellular >> filamentous (exhibiting 1D multicellular growth) >> planar/thalloid (exhibiting 2D multicellular growth) >> 3D forms that begin to resemble plants in their complexity. This is important b/c it is this evolutionary trend that gave rise to the Kingdom Plantae from amongst the green algae in particular.

Algae have been divided into groups based mainly on distinctive plastid accessory pigments. Because of these pigments, many groups of algae have been known by their color: hence red, green, brown, etc. Modern classifications delimit them with many other characters such as cell wall components, reproductive structures, and distinctive food storage molecules.

Algae are the dominant producers in aquatic environments, so they are very important ecologically. The materials available in lab today represent the major algal divisions, along with the cyanobacteria.

Some Useful Terms for Algae in this lab:

Bladder – see *pneumatocyst*.
Blade – the flattened analog of a leaf in many seaweeds.
Coenocytic – an organism whose cell(s) contain multiple nuclei (e.g., the alga Vaucheria exists as a long, sometimes branched filament that is just one or few giant cells).
Colonial – an "organism" actually made up of multiple organisms. In algae, these individual organisms are typically unicellular and they stick to one another by a special gelatinous matrix they secrete.
Eyespot – a patch of pigment on a membrane inside a cell that can detect light. These are typically red and found only in motile algae since the cell, once it detects light, can them swim towards it (or away from it if it is too much).
Filamentous – an organism that is long and thin (sometimes branching), and typically made up of a string of cells.
Holdfast – the branching structure at the base of many seaweeds that hold them to a substate.
Motile – to have directed mobility. Cells able to propel themselves by some means are said to be "motile." Usually this is achieved by way of a flagellum or two (e.g., *Chlamydomonas*). However, the filamentous cyanobacterium *Oscillatoria* can move by way of the entire filament and mass of filaments oscillating or twirling, and desmids such as *Closterium* can move in a somersaulting motion by secreting jets of mucilage through special pores in the cell wall alternatingly from different ends of the cell.
Pneumatocyst – an air-filled bladder like organ on many seaweeds that function for buoyancy to keep the blades afloat in a position to intercept sunlight.
Pyrenoids - centers of carbon dioxide fixation or starch synthesis within the chloroplasts of algae and hornworts, visible b/c of the density of enzymes in that region. Pyrenoids are not membrane-bound organelles, but specialized areas of the plastid that contain high levels enzymes such as ribulose-1,5-bisphosphate carboxylase/oxygenase (RubisCO).
Seaweed – a macroscopic marine alga. This term has no real technical definition and no taxonomic significance since some "seaweeds" are more closely related to unicellular or microscopic algae than they are to other "seaweeds".
Stipe – the stalk between the holdfast and the blades of many seaweeds.

A. Dichotomous Key to Freshwater Algae

Using the detachable key to freshwater algal genera on the last page of this lab, identify the 10 unknown monocultures of algae to their respective genera.

The algal samples in vials 1-10 are largely monocultures, but the presence of other species as contaminants is possible. Often there are small protozoa (microscopic animals) feeding on the algae in these vials, but do not be distracted by them. Focus your keying effort on the "majority" alga in each vial.

See instructor for the answers at the end of class. Important details are viewed at 200X and 400X (or higher) magnifications.

0. Working in pairs, make wet mounts (with coverslips) of two of the numbered algae tubes at a time from the back. Be sure to remember which number each slide is of – perhaps by number the slides with tape and pen.

  a. Do this by placing a drop onto slide (be sure to get some algae in it – you may have to flush the tube once with the pipette).
  b. Cover with coverslip (be careful laying the cover slip down so as not to destroy large colonial forms such as *Volvox)*.
  c. Do not let these wet mounts dry out until you are done looking at them.

1. Inspect the alga for color since this is a characteristic you'll often need to identify them.
Do this one (or both) of two ways:

  a. hold the tube up to the light or against white paper.
  b. then be sure to confirm this color again once you've got it under the scope since some of the tubes have soil contaminants in them and will make algae in the tube look brown when the cells are actually green, for example.

2. Work your way through the detachable key on the last page to identify the genus. You may visit section C in the lab manual with information about each algal group to confirm your tentative identification with pictures or descriptions.

3. Make drawings below and label with genus name, the magnification used for the drawing, and other structures such as **CHLOROPLAST(S), FLAGELLUM(FLAGELLA), PYRENOID(S), EYESPOT, NUCLEUS,** etc. Also record the color of your algae, as seen with the naked eye.

4. After making your identification, go to section C in the lab manual on the group and answer the questions.

1.	2.
3.	4.
5.	6.

7.	8.
9.	10.

B. OPTIONAL (per instructor choice- difficult to find algae November –March): Practical Algal Taxonomy

Taxonomy is the scientific discipline that deals with the formal naming, classification, and identification of organisms. A taxonomic key (which you used above) is a very useful tool that biologists and people who monitor water quality use to help identify which algae are found growing in the bodies of water they are interested in. Use the skills from Part B above to answer some of these practical questions.

1. What types of algae or cyanobacteria do grow in the Roddy Pond?

Additionally, do different species grow at different depths? It is conceivable that different algal species have different preferences in light intensity and temperature and we will test this hypothesis.

Sample from Roddy Pond the following (note that in winter months [november through March] most of the algae are low in the water column or on the bottom near the shore since they are not very active in photosynthesis).

1. Surface (the "pond scum")
2. Sample at 6 inches (18 cm) below the surface.
3. Sample deeper, between 3-5 feet (but do not scrape the pond bottom). You may need to sample from the pier for this deeper sample.
4. Place into glass jars/vials with lids and label.

5. Use the detachable dichotomous key to algae and cyanobacteria on the last page to identify samples of photosynthetic organisms taken from the pond.

2. Is Roddy Pond safe for cattle or cows to drink from?

Farmer's sometimes have major problems with cattle drinking from ponds with high densities of cyanobacteria (see the snippet of the 1999 article by Codd et al. on the next page). Although cyanobacteria resemble algae, they are bacteria and many cyanobacterial species make potent neurotoxins that can kill cattle. You can use the detachable dichotomous key to algae and cyanobacteria on the last page to identify samples of photosynthetic organisms taken from the pond. Pretend you are a farmer, and if you find cyanobacteria in great abundance, then you may have cause for concern.

European Journal of Phycology (1999), **34**:405-415 Cambridge University Press

Cyanobacterial toxins, exposure routes and human health

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Abstract

The production of potent toxins by bloom-, scum- and mat-forming cyanobacteria, in fresh-, brackish and marine waters, appears to be a global phenomenon. Cyanobacterial toxins can also be produced by cyanobacteria from terrestrial sources. The range and number of known cyanobacterial toxins are increasing apace as associated poisoning incidents are investigated, and increasingly powerful analytical methods are applied to complement toxicity-based studies on both natural samples and laboratory isolates of cyanobacteria. Water quality management to reduce toxic cyanobacterial mass developments, and schemes to mitigate the potential effects of cyanobacterial toxins, require an understanding of the occurrence and properties of the toxins and of the exposure routes via which the toxins present risks to health. Here, we review advances in the recognition of cyanobacterial toxins and their toxicity, and of the exposure routes with reference to human health, namely via skin contact, inhalation, haemodialysis and ingestion (the oral route).

(Received January 20 1999)
(Accepted May 11 1999)

C. Groups of Cyanobacteria & Algae

Regardless of whether or not you've seen living examples in lab today, read each section and answer the questions.

C1. Group 1: Cyanobacteria (blue-green algae)

Cyanobacteria are photosynthetic bacteria and were sometimes called "blue-green algae" by earlier botanists, but they are very different than the other organisms we typically call algae. That is, cyanobacteria are prokaryotes (i.e., they lack a nucleus and membrane-bound organelles). Whereas plants and other algae have chloroplasts in their cells to carry out photosynthesis, the whole cell of a cyanobacterium is homologous to a chloroplast. Like other prokaryotic cells, the cells of cyanobacteria are much smaller than eukaryotic cells. Although cyanobacteria have chlorophyll, their different mix of accessory pigments make them appear bluish-green. Many are unicellular, others are filamentous, while others are colonial.

1. Thought question: *If you were to have a filamentous (or unicellular) cyanobacterium and a filamentous (or unicellular) alga, how might you tell them apart? List at least two ways, based on your reading of the above paragraph.*

2. One of the unknown samples from section B was *Oscillatoria* or *Lyngbya*, which have filaments which can move or oscillate, causing the mass of filaments to move!

Did you see evidence for that?

What color were the masses of filament in the stock culture tubes or under the microscope?

3. *Which other genera of cyanobacteria did you find in the mystery monocultures in part A?*

Cross reference these answers to your drawings in part A above.

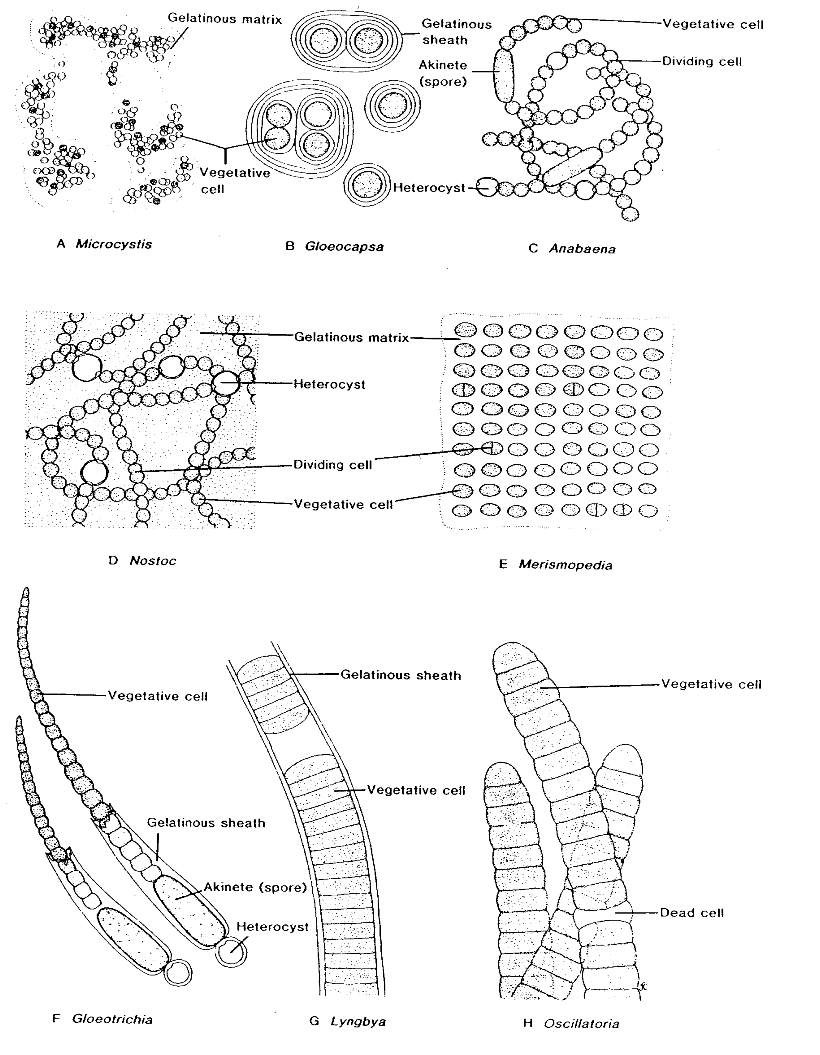

Figure at left: some cyanobacteria. A-B and D-E are more or less colonial, since the cells and/or filaments stick together in a gelatinous matrix.

<u>Above</u>: Cyanobacterial genera.

C2. Group 2: Dinoflagellates

Dinoflagellates are not bacteria but unicellular protists (i.e., in the Kingdom Protista according to a 5 or 6 kingdom classification scheme).

Thus, are dinoflagellates prokaryotes or eukaryotes?

We do not have dinoflagellates in any of the monocultures in lab today, but Roddy Pond sometimes has dinoflagellates in it (note: your particular lab may be too early in the season to see any). If there are any sampled in your pond water, you will know a dinoflagellate by its olive-brown color (due in part to accessory pigments PERIDININ or FUCOXANTHIN) and their elaborate shapes and ornamentation. Interesting phenomena in nature include bioluminescence and red tides caused by some marine species.

How many cells is each dinoflagellate?

Inspect your samples and the pictures below: How many flagella do these have and how is each arranged to help them move?

Read in your textbook about the "armor" or "plates"(p. 334). Are the plates inside or outside the cell(s)? What are they made of? How is this the same or different than a cell wall?

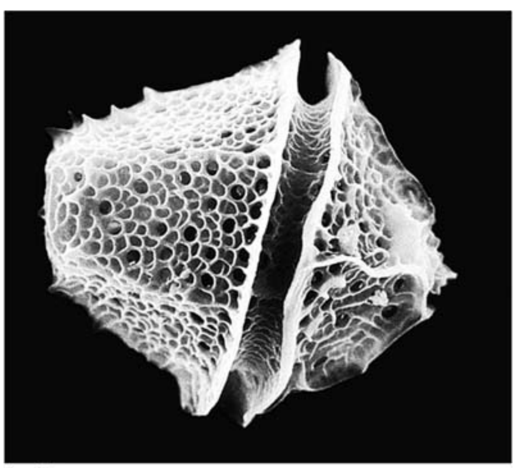

Figure 15-5c
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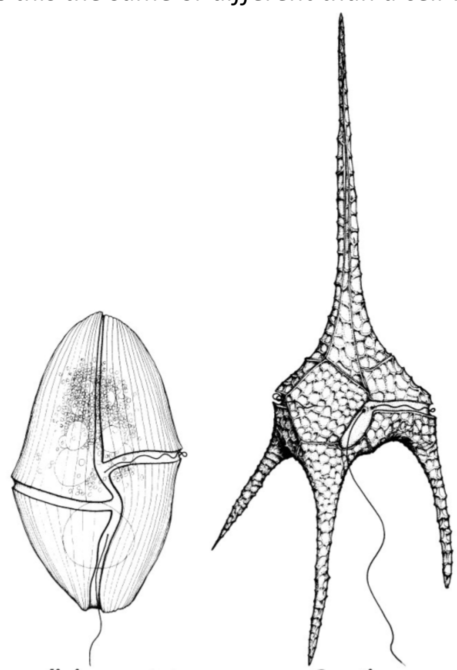

Figure 15-6
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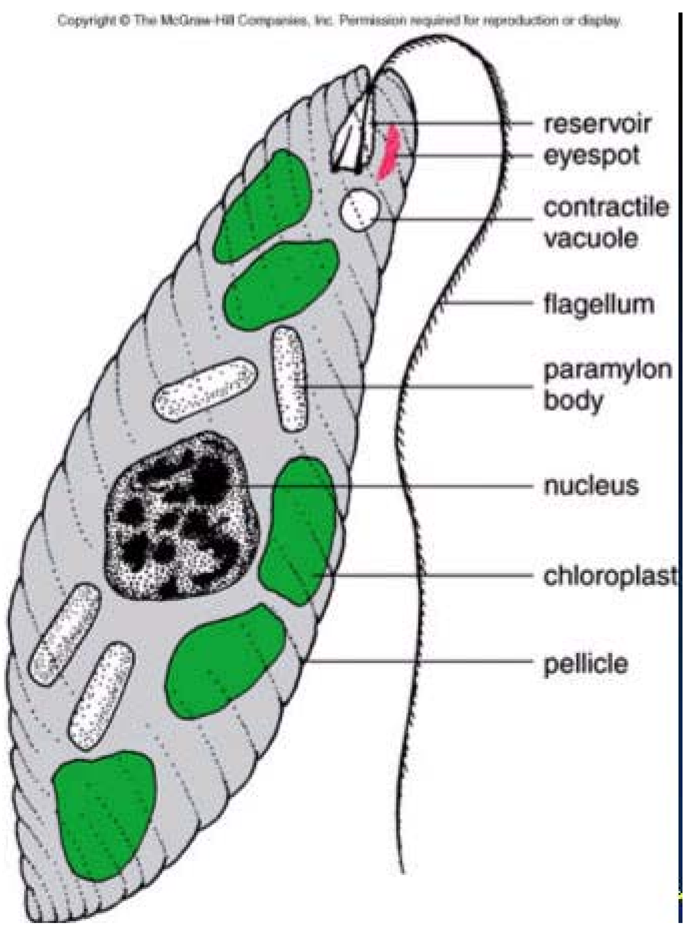
Figure above: Various forms of dinoflagellates.

C3. Group 3: Euglenoids

EUGLENA: PLANT OR ANIMAL? Remove a drop or two of the freshwater sample containing *Euglena gracilis* *Euglena gracilis* is just one of many (ca. 800-900) unicellular species of the genus *Euglena*. They do not have common names like birds or trees, so we have to refer to them by their scientific name *Euglena*.

Euglena are interesting because they combine superficial characteristics of both plants and animals. They can make their own food like a plant, through photosynthesis, but they can also eat other things, "like" an animal (although they actually eat more like an amoeba – through phagocytosis-- than an animal). They can also swim and move thanks to their flagellum. Scientists argued for years about which Kingdom to put them in: Animalia or Plantae? Right now they are in neither: according to some classifications they are in the Kingdom Protista with other simple (often microscopic) eukaryotes, such as amoeba and paramecium. Protists are eukaryotes, typically small or microscopic, which lack sufficient characteristics to otherwise justify their inclusion in the animal, plant, or fungal kingdoms.

A euglena's body is unicellular. The outermost envelope is the plasma (cell) membrane, beneath which lies the pellicle (a matrix of protein bands that spiral down the length of the cell). The pellicle helps the euglena maintain its distinctive shape, although it is quite flexible and can be flexed by the cell in an inch-worm type fashion. Euglena typically store their excess carbohydrates as paramylon (a polymer of glucose related to starch), distributed throughout the cytoplasm as "paramylon bodies." Their chloroplast pigment profile is similar to that of green algae and plants: chlorophyll a, b, and carotenoids. At one end of a cell is a contractile vacuole, which helps expel excess water absorbed by the cell from its freshwater environment.



Sometimes, since they live in water, if there are millions of euglena together, they form a mat on the surface of a pond or marsh that you can see. It looks slimy, a lot like algae. Some people say it looks like "pea soup". Euglena get into swimming pools too, if they are not cleaned regularly.

1. Are euglena unicellular or multicellular?

2. Are euglena animals, plants, protists, or bacteria? Prokaryotic or eukaryotic?

3. What organelle carries out photosynthesis?

4. Are euglena autotrophic or heterotrophic? Explain.

5. How many flagella did the euglena from the unknown algal cultures above have?

6. Does the flagellum or flagella work by "pulling" or "pushing" the cell through the water?

7. What is the eyespot used for?

8. What is the function of the nucleus?

10. What is the function of the contractile vacuole? What would happen if the cell did not have this organelle.

11. Did the shape of Euglena cells change while you observed them? Or were they rigid?

C4. Group 4: Brown, Golden-brown, and Yellow-brown Algae .

A diverse group united by the accessory pigment fucoxanthin and carbohydrate reserves laminarin or the very similar chrysolaminarin.

C4.1. Diatoms

Diatoms are unicellular or colonial organisms that are exceedingly important components of phytoplankton of both marine and freshwater systems. In addition to their chlorophyll, diatom chloroplasts contain the accessory pigment fucoxanthin, and the chlorophyll plus fucoxanthin makes them appear golden-brown or brown-yellow in color. Their carbohydrate reserves are in the form of chrysolaminarin, a starch relative. Fossil evidence indicates that they evolved during or before the Jurassic. Fossil beds of diatoms are often called "diatomaceous earth," which is often used by humans as a water filter matrix (e.g., pool water filters are often diatomaceous earth), or as a polishing agent. In the station in the back, we have a sample of diatomaceous earth (i.e., diatom fossils). Much of our oil deposits in world are the transformed organic remains of diatoms and other marine algae.

Diatoms lack flagella and are non-motile. Outside of the plasma membrane lies the cell wall, which consists of two halves made of hydrated silica dioxide. The cell wall of diatoms is called a frustule and the two halves fit together like a Petri dish. These frustules are very ornate and these ornamentations are used to distinguish species. Biotechnologists have begun to look at manipulating diatoms (through artificial selection) to produce nanotech valves for nanoscale drug delivery, etc.

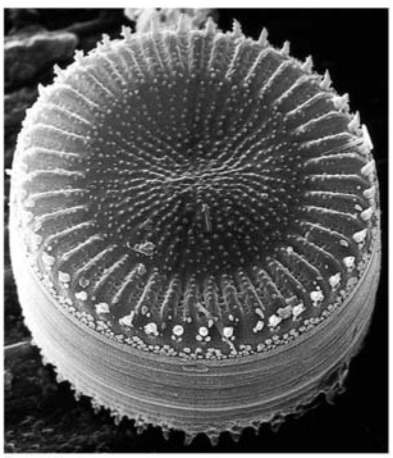


Figure 15-20d
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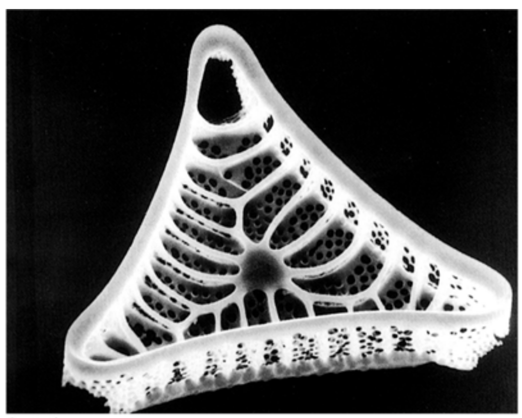

Figure 15-20b
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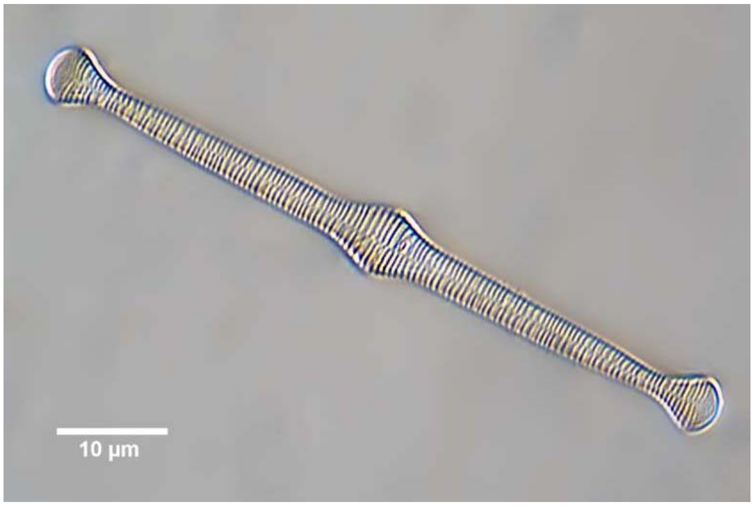
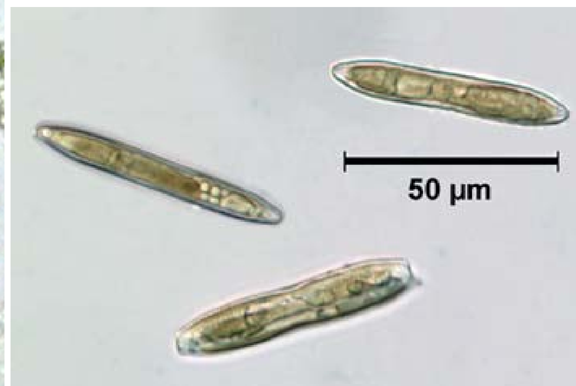
Above, scanning electron micrographs of a whole diatom (left) and the half of a frustule, showing the inside face (right).



Above, *Tabellaria* from a freshwater pond.



Above: *Navicula* (left) and *Synedra* (right). (*http://biology.missouristate.edu/phycology*)

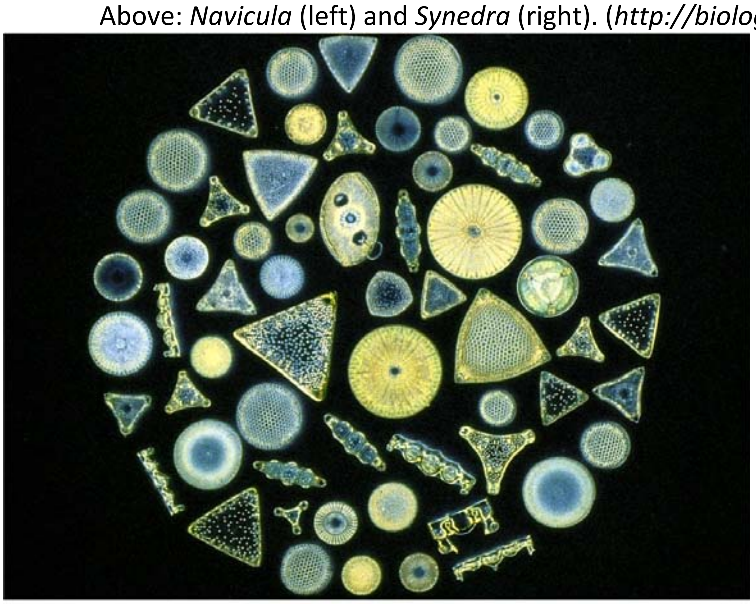


Figure 15-20a
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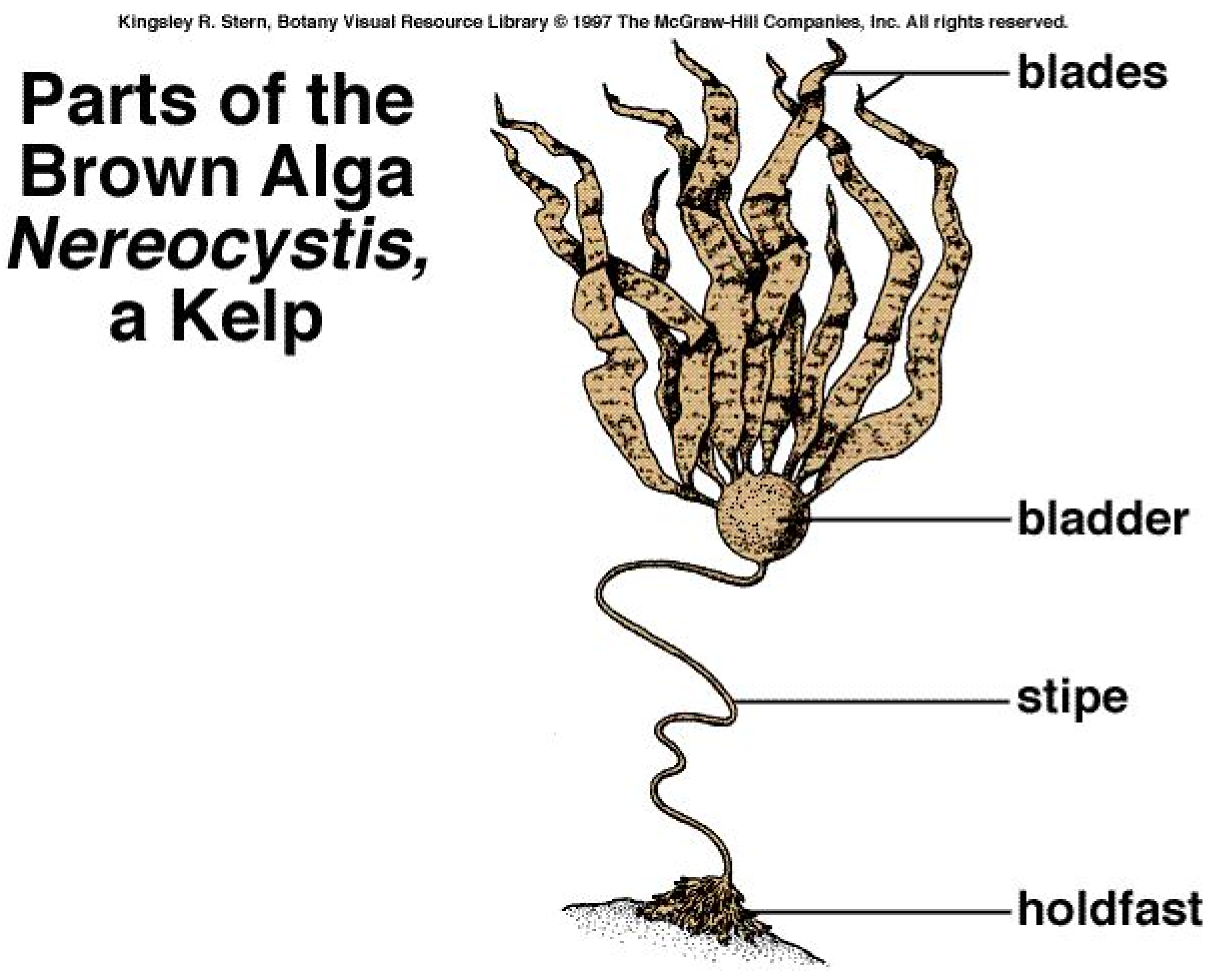
Fig. 15-20a from Raven et al. (2007). An artful representation of many different types of diatoms.

C4.2. Brown Algae

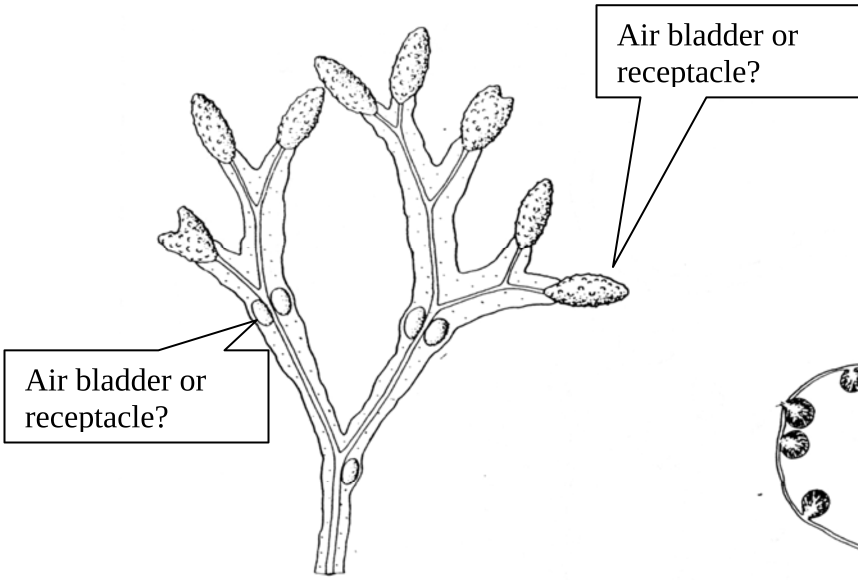
When marine algae get big (macroscopic), they are called <u>seaweeds</u>. Most browns are relatively big, and all are multicellular. Only 6 of 265 genera occur in freshwater. Although they have chlorophyll, their special carotenoid <u>fucoxanthin</u> (which they share with diatoms and many dinoflagellates) impart a brown color to them. These algae store excess carbohydrates in the form of <u>laminarin</u>, a relative of starch that is very similar to <u>chrysolaminarin</u> of diatoms. Brown algae have cellulosic cell walls.

Well known members include *Fucus* (rockweed), *Laminaria* and *Macrocystis* (the kelps).

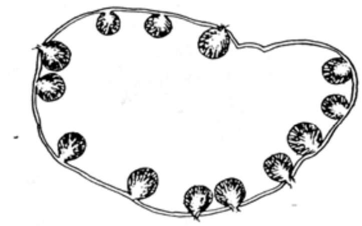
Common structures will include a <u>HOLDFAST</u>, a <u>STIPE</u>, A <u>BLADDER</u> (for buoyancy; aka <u>pneumatocyst</u>), and <u>BLADES</u> to function like "leaves".



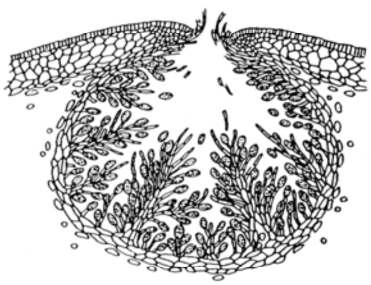
Holdfast Logic Question: *In what way(s) do holdfasts resemble roots, and in which way(s) do they not? HINT: think about function(s).*



A. *Fucus*, plant with receptacles and air bladders.

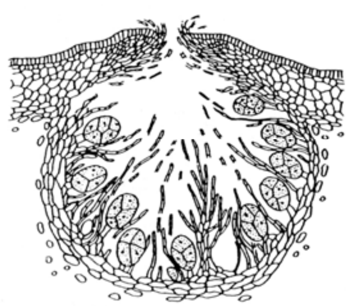


B. x-section of receptacle and smaller conceptacles.

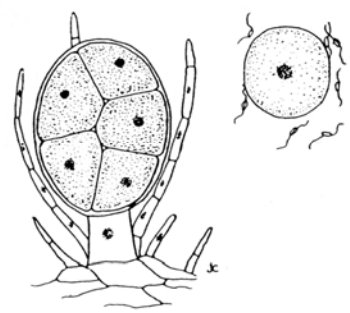


C. Male conceptacle with antheridia.

D. Antheridia with sperm.



E. Female conceptacle with oogonia.



F. Oogonium with eggs.

Above: *Fucus*, illustrations of the plant, as well as the reproductive parts.

C4.5. Group 5: Glaucophytes

Extremely small group of algae closely related to red and green algae. Will not be covered in lab.

C4.6. Group 6: Red Algae

The red algae are largely marine and multicellular "seaweeds". A class of accessory pigments (phycobilins) makes them reddish. Red algae store carbohydrates as floridean starch, a starch relative. Red algae have cellulosic cell walls.

We don't have any living red algae in the lab. There are herbarium specimens for your observations, however.

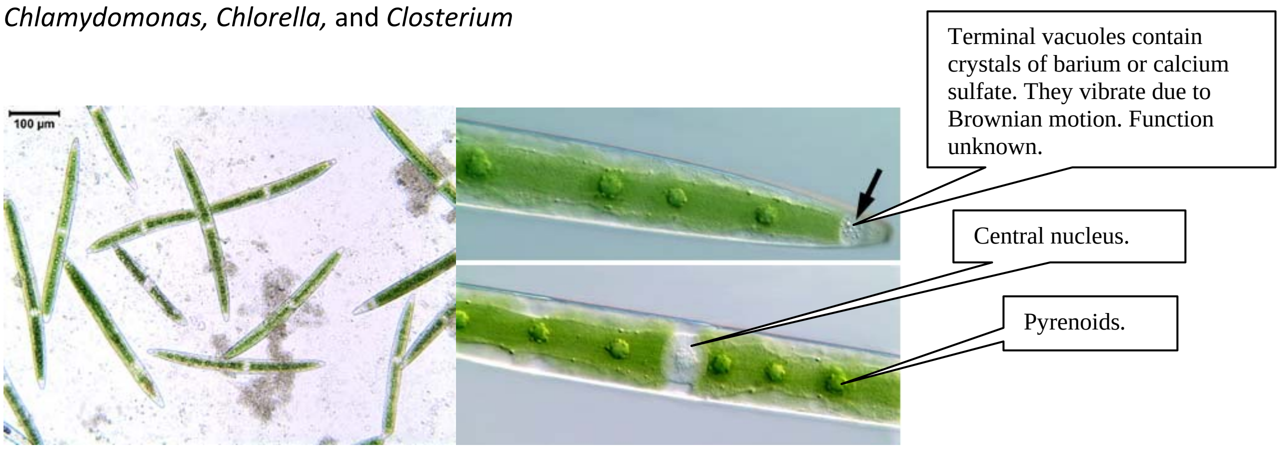
C4.7. Group 7: Green Algae

The green algae are a very large and diverse group. They range from unicellular organisms such as *Chlamydomonas* or *Chlorella* to colonial organisms such as *Volvox*, to mulicellular organisms such as *Ulva* (sea-lettuce) and *Chara* (stonewort). Green algae share with plants the same chloroplast pigment profile (chlorophyll a, b, and the carotenoids beta-carotene and xanthophylls). When a cell wall is present, it is cellulosic.

Many of the unlabeled monocultures of algae in the lab were green algae. Presumably, you have now identified them all.

C4.7A. Unicellular forms: all three of the algae below were represented in the unknown algal cultures. Which numbers were which?

Chlamydomonas, Chlorella, and *Closterium*



Above: Closterium. This unicell is bilaterally symmetric, with two equal halves and a single chloroplast in each half; such unicellular algae with symmetrical halves are called "desmids." Like many desmids, *Closterium* can move in a somersaulting manner by secreting mucilage from alternating ends of the cell through special pores in the cell wall. (http://silicasecchidisk.conncoll.edu/LucidKeys/Carolina_Key/html/Closterium_Main.html)

Which of the algae above is an indicator of polluted fresh or salt waters? See the poster in the back or side of the room.

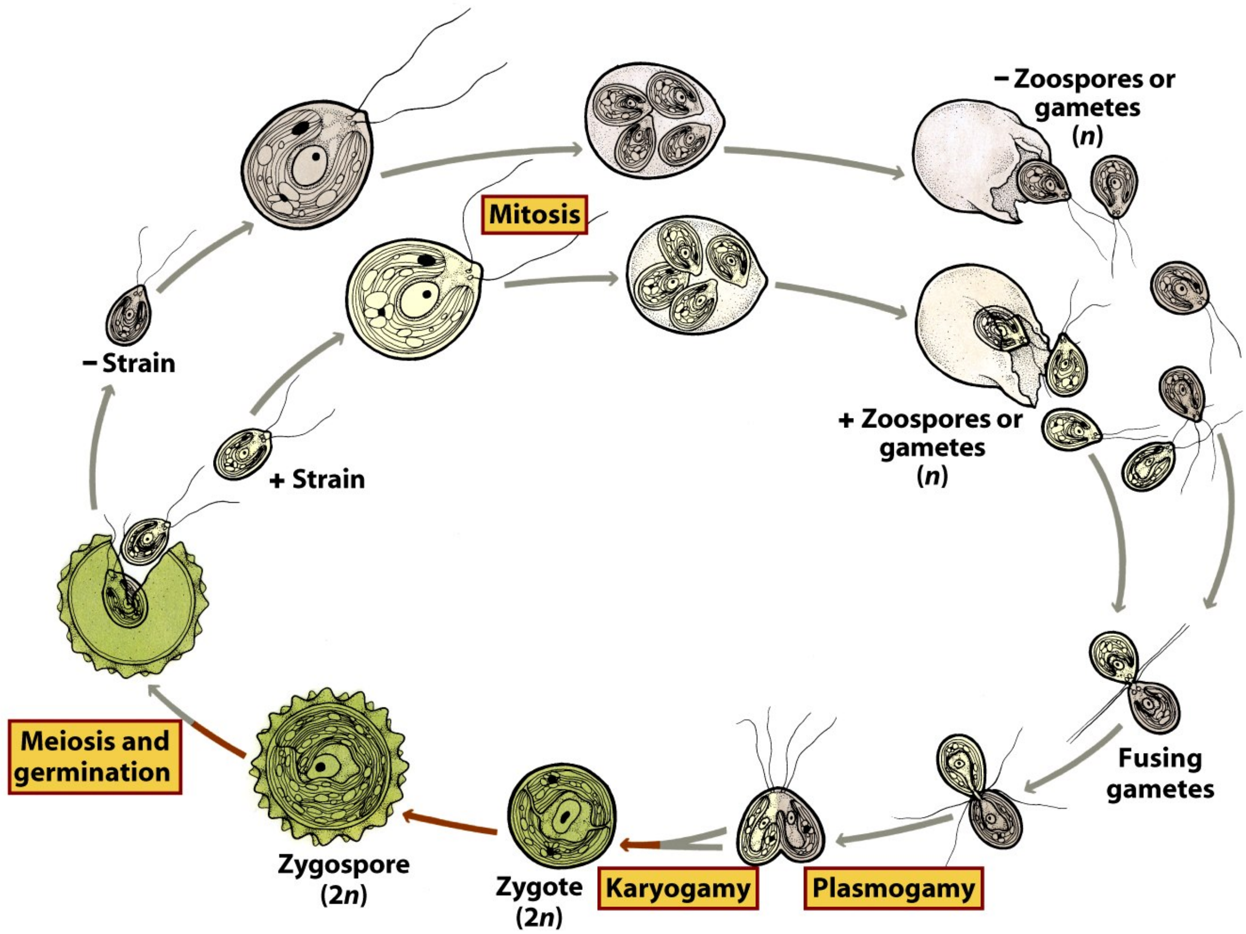


Figure 15-41
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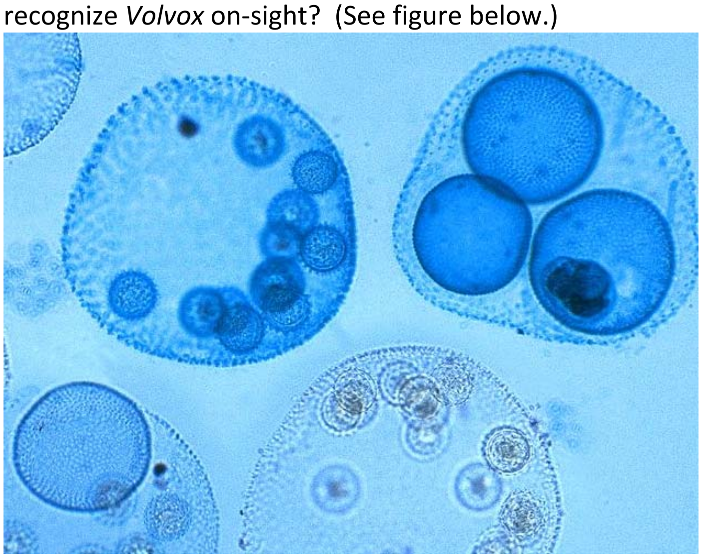
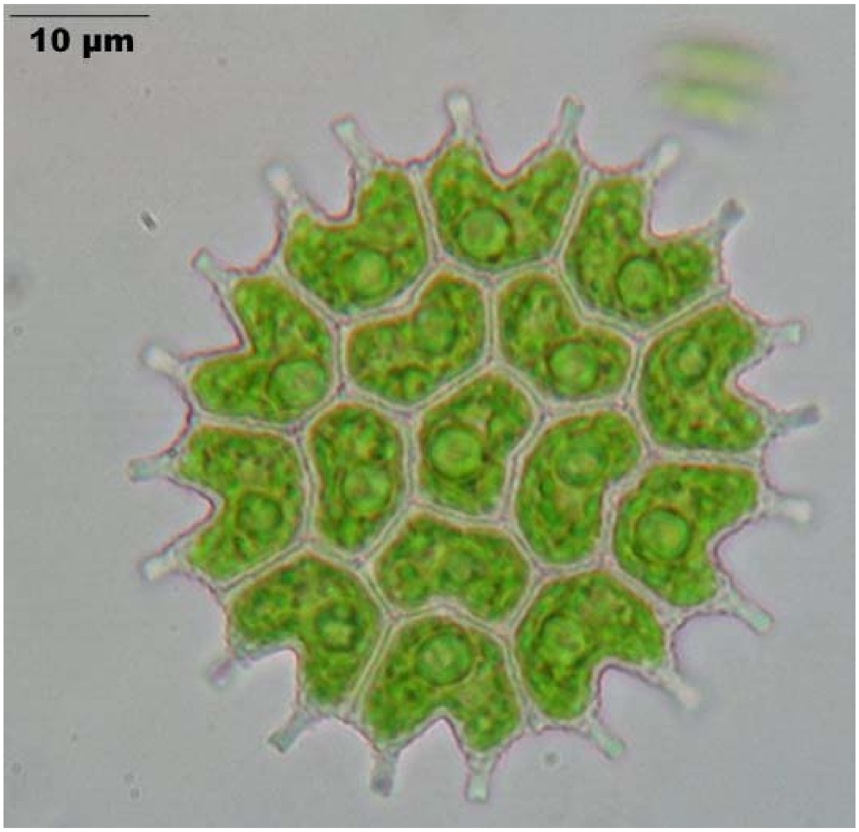
Above: Life cycle of *Chlamydomonas*, a unicellular green alga.

C4.7B. Colonial forms: e.g., *Volvox* & *Pediastrum* (sometimes found in the pond). Can you recognize *Volvox* on-sight? (See figure below.)



Above: *Volvox* with daughter colonies inside larger parent colonies.



Above: *Pediastrum*, a plate-like colonial freshwater (pond) alga.

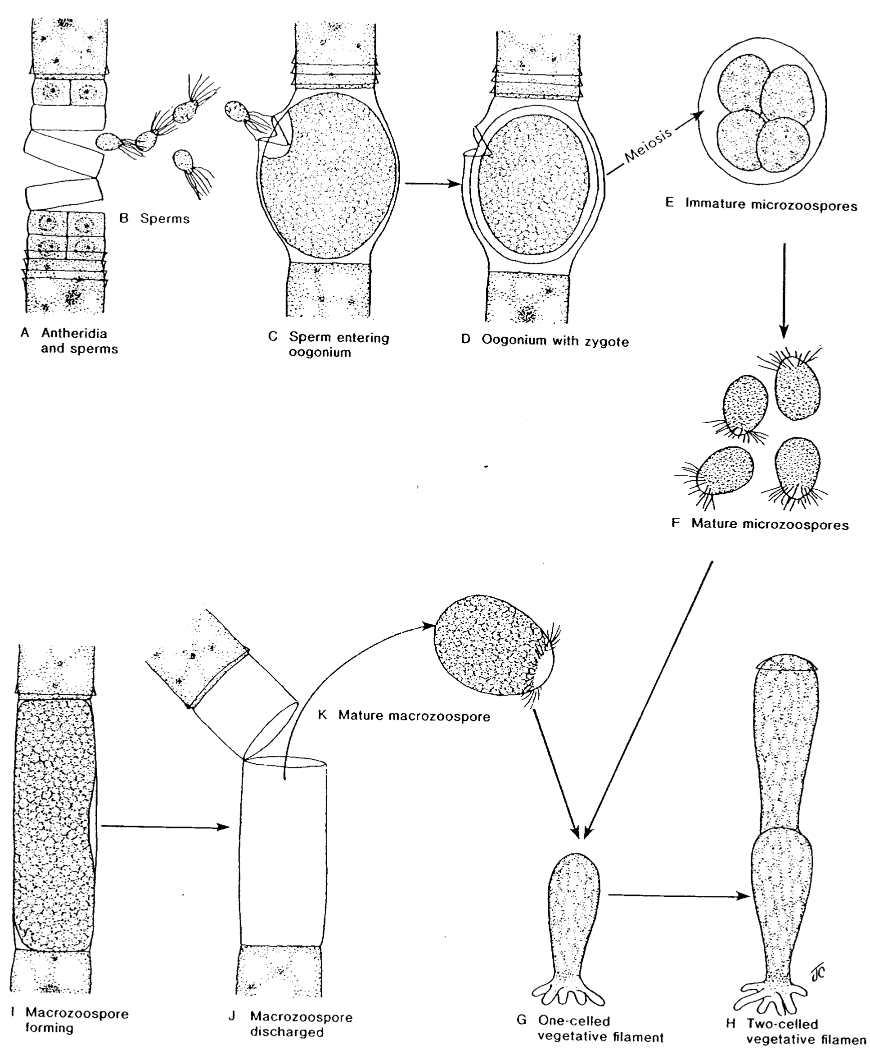
C4.7C. Filamentous forms: *Spirogyra* was represented in the algal cultures and possibly the pond water. How about *Oedogonium*?

Why is Spirogyra named as such?

Can you spot pyrenoids (protein-rich sites of carbon fixation and starch synthesis) in Spirogyra chloroplasts?

How do you distinguish these two genera (look at the key or the figure below)?

Figure 23.2 *Oedogonium*



Above, life-cycle of the green alga *Oedogonium*.

C4.7D. Macroscopic (seaweed) forms: see pictures, posters and herbarium specimens at stations around the periphery of the room.

Dichotomous Key to Some Freshwater Algal Genera (© 2010, MU Botany)

1. Alga unicellular and cells round, ovoid, or elliptic (cells may be clumped together but do not confuse this with a colony- which should be regular and ordered- or a filament)........................2
1. Alga colonial or filamentous (if colonial, then cells regularly and orderly grouped into spherical clusters or clusters of 2-4 cells).. 7

2. Cells light golden-brown with highly transparent glass-like cell walls, shape elliptic (football-shaped) or long, thin and bone-shaped or rectangular, non-motile (Diatoms).................................... 3
2. Cells bright-green; round, ovoid, or slightly crescent-shaped; motile or non-motile...................................... 4

3. Cells elliptic (football-shaped) or oblong (ends tapering)....................... Synedra (Diatom)
3. Cells rectangular or bone-shaped (ends blunt or bulbous)..................... Tabellaria (Diatom)

4. Cells narrow (longer than wide, although sometimes changing shape).. 5
4. Cells rounder or more ovoid.. 6

5. Cells apparently non-motile; very thin, with pointy ends, slightly crescent-shaped, with two equal halves – the nucleus in the middle and each half containing one chloroplast.................................. Closterium (Desmid Green Alga)
5. Cells motile; not so thin and with rounded ends; patch of red pigment near flagellum insertion.. Euglena (Euglenoid)

6. Cells ovoid, usually rather motile, with a groove on the narrower end where two flagella insert; red eye-spot present... Chlamydomonas (Green Alga)
6. Cells round, not apparently motile (not flagellate), no red eye............ Chlorella (Green Alga)

7. Alga colonial - cells grouped into groups of 2-4 cells or globular clusters, & held together by gelatinous matrix... 8
7. Alga a filament, branched or not.. 9

8. Colonies of 2-4 cells, cells blue-green.. Eucapsis (Cyanobacterium)
8. Colonies round, some cells golden-brown-green................................... Synura (Golden-Brown Alga)

9. Cells blue-green (Cyanobacteria).. 10
9. Cells or chloroplasts green or yellow-green... 12

10. Filaments unbranched and straight or arching for long stretches........ 11
10. Filaments branched frequently and noticeably curved......................... Tolypothrix (Cyanobacterium)

11. Filaments with constrictions between the cells.................................... Anabaena (Cyanobacterium)
11. Filaments without constrictions between the cells; moving.............. Oscillatoria (Cyanobacterium)

12. Filament branched and apparently coenocytic (all one cell or very few cells – that is, with few or no internal partitions)....................... Vaucheria (Yellow-Green Alga)
12. Filament obviously multicellular.. 13

13. Chloroplast(s) in each cell not round, instead somewhat coiled.. Spirogyra (Green Alga)
13. Chloroplast(s) round... Oedogonium (Green Alga)

Dichotomous Key to Some Freshwater Algal Genera (© 2010, MU Botany)

1. Alga unicellular and cells round, ovoid, or elliptic (cells may be clumped together but do not confuse this with a colony- which should be regular and ordered- or a filament)........................2
1. Alga colonial or filamentous (if colonial, then cells regularly and orderly grouped into spherical clusters or clusters of 2-4 cells).. 7

2. Cells light golden-brown with highly transparent glass-like cell walls, shape elliptic (football-shaped) or long, thin and bone-shaped or rectangular, non-motile (Diatoms).................................... 3
2. Cells bright-green; round, ovoid, or slightly crescent-shaped; motile or non-motile...................................... 4

3. Cells elliptic (football-shaped) or oblong (ends tapering)....................... Synedra (Diatom)
3. Cells rectangular or bone-shaped (ends blunt or bulbous)..................... Tabellaria (Diatom)

4. Cells narrow (longer than wide, although sometimes changing shape).. 5
4. Cells rounder or more ovoid.. 6

5. Cells apparently non-motile; very thin, with pointy ends, slightly crescent-shaped, with two equal halves – the nucleus in the middle and each half containing one chloroplast.................................. Closterium (Desmid Green Alga)
5. Cells motile; not so thin and with rounded ends; patch of red pigment near flagellum insertion.. Euglena (Euglenoid)

6. Cells ovoid, usually rather motile, with a groove on the narrower end where two flagella insert; red eye-spot present.. Chlamydomonas (Green Alga)
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