Biology of Algae

Laboratory Guide

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BSC 464/564 • The University of Alabama • Spring 2013

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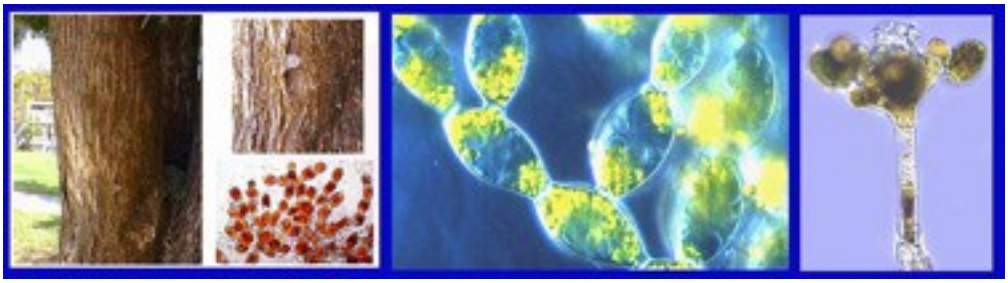
Introduction

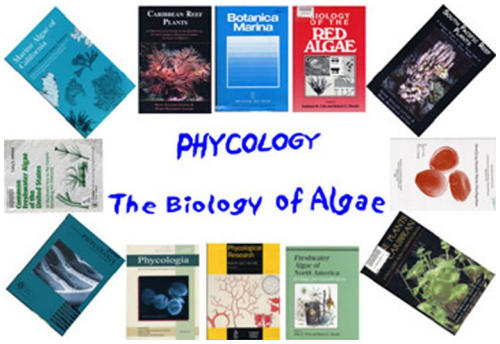


Examples of subaerial algae growing in our backyards

This Biology of Algae Laboratory Guide has been designed to complement lecture materials for BSC464/564 in order to give you a better understanding of the biology of these organisms. Fieldtrips, algal collections, isolation and culturing, fixation and preservation, identification, herbarium curation, and other activities are included during this course. These activities will provide you with the opportunity of having hands-on-experience while studying algal samples. During these lab sessions we will gain familiarity with methods to collect and preserve algae for scientific study. Throughout the semester we will review common examples of algae. These organisms are usually found in habitats readily available in the Southeastern USA. Each algal group will be analyzed in terms of their biodiversity, vegetative and reproductive morphologies, as well as its relationship to human affairs. These activities will prepare you toward the end of the semester, as you become familiar with the technical terms and algal appearance, to focus on the Identification of algal specimens. An algal collection should be presented for grading along with a systematic list and field reports during the final week of classes.

Field trips are organized to visit a diverse set of habitats: marine, freshwater, and terrestrial. A two-day trip to Dauphin Island is scheduled to collect and study intertidal seaweeds and marine phytoplankton. Other areas will be visited to compare terrestrial, lotic and lentic environments. Seaweeds, freshwater algae, phytoplankton and terrestrial algae will be collected during these fieldtrips. A short visit to Marrs' spring on UA campus will provide you with the opportunity to practice the techniques for collecting algae. During these fieldtrips is recommended to bring flashlights, field notebooks, a field knife, a hand magnifier, insect repellent, sun protection, and proper clothing. Check your class syllabus for specific dates and back-up dates for fieldtrips.

Guidelines



Lab reports

At the end of each lab session you will turn in your report for grading. Observations must be made on non-ruled paper and kept in a binder with your name. Use only pencils and one side of paper (no pens allowed in your report). Remember that a good graphic representation of an organism is not indicative of art talent, but rather good observations. Each drawing should be correctly labeled, including:

Scientific name (i.e. *Chlamydomonas*)
Classification information (class, order, and family)
Part being observed (i.e. akinete)
Other features (i.e. flagella, pyrenoid, cell wall, etc)
Magnification (eyepiece magnification *times* objective magnification; i.e. 100x)

Materials to bring to each laboratory session:

- A letter size three-ring binder
- Algae textbooks
- HB pencils
- Twin tip (fine/ultra fine points) Sharpie black
- Slides kit: available from the supply store
- Pencil sharpener
- Eraser

General laboratory Procedures

1. Each session will start with an organizational discussion. Check the board for the agenda. Laboratory session starts at l PM sharp, be on time for general directions and instructions. There is no make up for missed lab sessions.

2. At the end of each lab session you will turn in your report for grading. Make sure you report has been completed with all the requirements (answers, drawings, labels, taxonomy, correct spelling, writing your name on the report, data, etc).

3. Treat microscopes well: Hold the microscope with both hands when you pick it up from the cabinet and after you finish your lab work. Clean the lenses only with lens paper, before and after your lab work. Learn to adjust microscopes: at the beginning of your session, at every magnification, and at the end of your lab work. Do not leave any slides in the microscope! If you are found mistreating a microscope you will be asked to do extra work (microscope use demo in front of the class).

4. Re-use the microscope slides and coverslips. After using your microscope slides and coverslips, wash them with water and dry them (carefully) with a paper towel. Broken microscope slides and coverslips should be disposed in the box marked as "Broken Glass" never in the regular trash bin.

5. We will use stains in a regular basis (i.e., Lugol solution, cresyl blue, etc.) be careful with your clothing. Stains from these dyes are permanent.

6. Be considered to yourself and others: clean your work area before and after your lab session.

7. Return all materials (prepared slides, dissecting needles, etc) to the demo area after you finish your work.

8. Protect yourself and others: we will be using some sharp objects (dissecting needles, blades) and harsh chemicals (alcohol, stains), practice common sense and read all posted warnings, be safe.

Organization for laboratory and fieldtrips

Lab sessions:

1. Cyanobacteria
2. Chlorophyta I: Ulvophyceans
3. Chlorophyta II: Chlorophyceans and Euglenoids
4. Rhodophyta I: Bangiophyceans
5. Rhodophyta II: Florideans
6. Cryptophyta, Haptophyta, and Dinophyta

7. Stramenopiles I: Diatoms-Tribophyceans!
8. Stramenopiles II: Phaeophyceans

Algal Identifications sessions:

9. Algal Identifications I
10. Algal Identifications II
11. Algal Identifications III
12. Algal Identifications IV
13. Algal Identifications V

Fieldwork sessions:

14. UA Campus Fieldtrip
15. Lake Lurleen Fieldtrip
16. Dauphin Island Sea Lab Fieldtrip

List of materials needed for algal collections during fieldtrips

a) Pencils
b) Twin tip (fine/ultra fine points) Sharpie black
c) Ziploc bags, several sizes
d) Vials with screw caps, several sizes (save them at home)
e) Scrapper, check local hardware stores
f) Small plastic bucket
g) Home-made herbarium plant press: 2 straps and 2 12X16" hardboards

Graduate Teaching Assistant

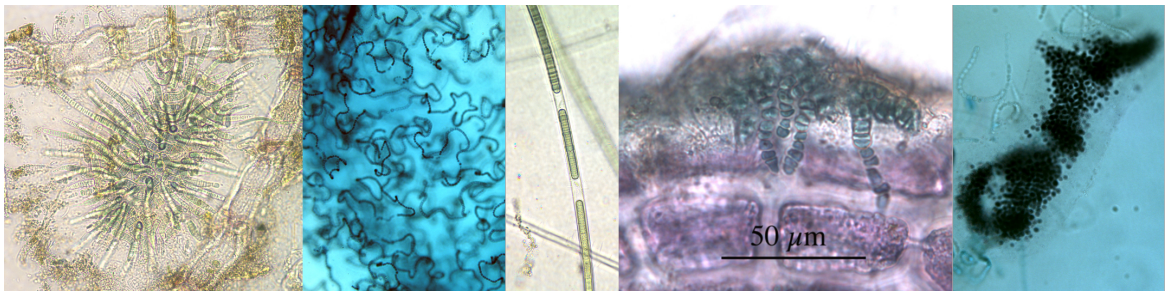
Name:
e-mail:
Phone:
Office:
Office hours:

Facebook page group

Algae Study Group

Cyanobacteria

THE BLUE-GREEN ALGAE



Examples of blue green algae: Calothrix, Nostoc, Lyngbya, Entophysalis and Microcystis.

Introduction

The Cyanobacteria or blue green algae have unicellular, colonial or filamentous thalli. A cell wall is present and made of peptidoglycans, similar to that of non-photosynthetic bacteria. Their prokaryotic cells are pigmented in the periphery of the cell (chromatoplasm region) with a blue-green color due to chlorophyll *a*, phycobilins, and carotenoids The colorless central area (centroplasm region) contains the genetic information (DNA). No plastids or other organelles such as nucleus, mitochondria, or flagella are present. There are several types of granules and some of them are storing chemical products such as the cyanophycin and glycogen (cyanophytan starch) granules. Asexual reproduction is common while sexual reproduction is absent.

The current classification of cyanobacteria is controversial with molecular and morphological data in disagreement.

Classification. The main morphological groups are:

- GROUP I: Unicellular and colonial, lacking specialized cells or reproduction
- GROUP II: Filaments lacking specialized cells, spores, heterocysts, or akinetes
- GROUP III: Producing exospores
- GROUP IV: Producing endospores
- GROUP V: Cyanobacteria with heterocysts and akinetes
- GROUP VI: Filaments with true branching

Biological material for examination

1) *Anabaena* (free and endophytic)
2) *Fischerella*
3) *Gloeocapsa*
4) *Gloeotrichia*
5) *Lyngbya*
6) *Microcystis*
7) *Nostoc*
8) *Oscillatoria*
9) *Spirulina*
10) *Tolypothrix*

Procedure

Prepare microscope slides with the cyanobacteria. Observe your samples with lower magnification first and then proceed at higher magnification. Make your observations using pencils

1. In unicellular and colonial thalli, label the cell wall, chromatoplasm, centroplasm, granules, and mucilage

2. In filamentous forms, look for cells, trichome, filament, sheath, heterocyst, akinetes, hormogonia, necridia, pseudo-branching, heterotrichous, true-branching, and polarity.

Results

1. Name the genera found with the following morphologies:
    a. Unicellular forms:
    b. Colonial thalli:
    c. Trichome:
    d. Filament forms:
    e. Uniseriate filaments:
    f. Multiseriate filaments:
    g. Sheaths:
    h. Heterocysts:
    i. Akinetes:
    j. Polarity:
    k. Hormogonia:
    l. Necridia:
    m. Simple (unbranched) filaments:
    n. False branching filaments:
    o. True branching filaments:
    p. Heterotrichous filaments:

2. Cyanobacteria with basal heterocysts?

3. Cyanobacteria with intercalary heterocysts?

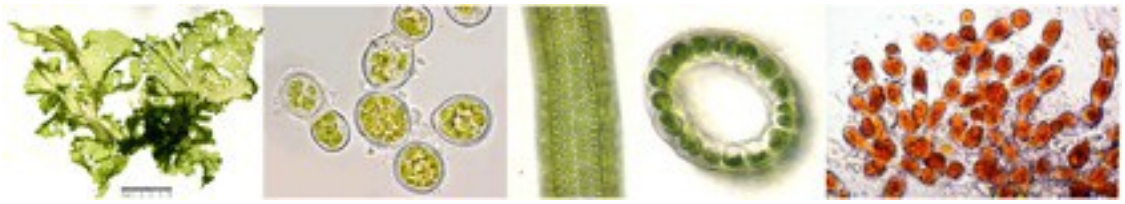
4. What is the function of the hormogonium?

5. Cyanobacteria living as endophytes in aquatic ferns:

6. What are the benefits for each participant in the relationship *Azolla-Anabaena*?

Chlorophyta I

THE GREEN ALGAE: ULVOPHYCEANS



Examples of ulvophycean green algae from marine and terrestrial environments

Introduction

The green algae or Chlorophyta are a diverse group with a wide range of morphologies. Unicellular, colonial, filamentous and parenchymatous thalli are some examples. The green color is predominantly chlorophyll a and b as well as several xanthophylls and carotenes. Starch is the storage product, and similar to that of the seed plants. Flagellated cells are usually isokont (similar in shape), acrokont (anteriorly attached), and acronematic (smooth); although in some cases can be stephanokont (forming a ring). Although often conspicuous in the marine environments, green algae are far more diverse in freshwater habitats.

Classification of Chlorophyta

- CLASS "PRASINOPHYCEANS". Primitive, marine, and unicellular green algae. They represent the early divergent green algae. Not a "natural" or monophyletic group.
- CLASS ULVOPHYCEAE. The marine greens and the terrestrial Trentepohliales.
- CLASS CHLOROPHYCEAE. The freshwater green algal celebrities.
- CLASS TREBOUXIOPHYCEAE. The symbiotic green algae, or "fungal" lovers
- CLASS CHAROPHYCEAE. The "conquerors" green algae and ancestors of the land plants.

Biological Materials for Examination

PHYLUM CHLOROPHYTA

CLASS Prasinophytes

Platymonas (=Tetraselmis)

CLASS Ulvophyceae

Order Ulotrichales

Ulothrix

Order Ulvales

Entermorpha

Ulva

Order Cladophorales/Siphonocladales

Cladophora

Chaetomorpha

Order Trentepohliales

Trentepohlia

Cephaleuros

Order Caulerpales

Caulerpa

Bryopsis

Codium

Order Dasycladales

Acetabularia

Procedure

Prepare microscope slides with the green algae as instructed. To observe pyrenoids add a drop of Lugol Solution (IKI). Observe your samples at lower magnification first then proceed with higher magnification.

1. With unicellular forms label cell wall, plastid, pyrenoid, and flagella.

2. *Ulothrix, Cladophora, Chaetomorpha, Trentepohlia, Cephaleuros,* and *Bryops*is: label cell wall, plastid, pyrenoid, cross walls, and type of branching (if any).

3. *Ulva, Entermorpha, Codium* and *Caulerpa:* Make observations of the macroscopic thallus and label holdfast and erect portion (blade, tubular, etc). With a razor blade make thin cross sections and prepare wet mounts.

    a. *Ulva*: Observe the distribution of cells in cross section, plastids, and pyrenoids

    b. *Enteromorpha*: Observe the distribution of cells in cross section, plastids, and pyrenoids

    c. *Codium*: Observe the pseudoparenchymatic and coenocytic nature of the thallus, utricles, and gametangia

    d. *Caulerpa* spp: Observe the coenocytic thallus and label stolons, rhizoids and blades. In a cross section observe at the microscope the trabeculae and plastids

4. In *Acetabularia* label holdfast, pedicel, and whorl of rays. At the microscope observe gametangial rays and cysts.

Results

1. Name the genera found with the following:
    a. Unicellular forms:
    b. Filament forms:
    c. Uninucleated forms:
    d. Multinucleated forms:
    e. Siphonous forms:
    f. Discoid thallus:
    g. Subaerial forms:
    h. Pseudoparenchymatous thallus:
    i. Laminar/blade thallus:
    j. Tubular thallus:
    k. Trabeculae:
    l. Cysts:
    m. Utricles:
    n. Heteroplastids:

o. Parietal plastids:

p. Reticulated plastids:

2. What is the function of a pyrenoid?

3. What are the cell (uni-, multinucleate) and thallus (simple, branched, uniseriated, coenocytic) differences among the following taxa:

	Ulothrix	*Cladophora*	*Trentepohlia*	*Chaetomorpha*	*Bryopsis*
Cells (uni- or multi-nucleated)					
Thallus (simple, branched, uniseriated, coenocytic)					

4. What is the advantage of a subcuticular thallus in a terrestrial environment?

5. What is the function of stolons in *Caulerpa*?

6. What is the function of cysts in *Acetabularia*?

7. Define the types of life cycles of

Ulothrix:

Cladophora:

Codium:

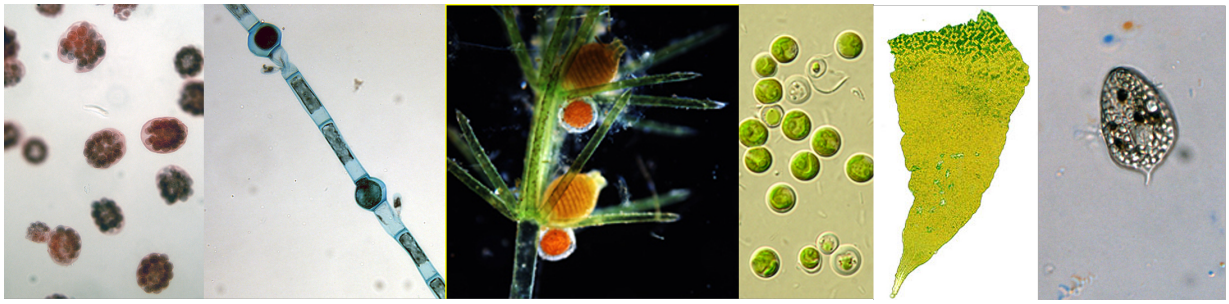
Acetabularia:

8. Compare the Ulvophycean orders based on morphology:

	Cells (Uni or multinucleated)	**Thallus** (simple, branched, uniseriated, coenocytic)	**Life Cycle** (sporic, gametic, zygotic; iso- or heteromorphic)
Ulotrichales			
Ulvales			
Cladophorales			
Trentepohliales			
Caulerpales			
Dasycladales			

Chlorophyta II

THE GREEN ALGAE: CHLOROPHYCEAE, TREBOUXIOPHYCEAE, AND CHAROPHYCEAE; THE EUGLENOIDS



Examples of other green algae *(Pleodorina, Oedogonium, Chara, Trebouxia, Prasiola)* and euglenoids (*Phacus*).

Introduction

This is the second part of the green algal survey. During this session we will be studying representatives of the Chlorophyceae, Trebouxiophyceae, and Charophyceae. We will also observe some euglenoides. Although euglenoids are not formally "green algae" they are descendents, via secondarya endosymbiosis, from an ancestral green alga (plastid) and an heterotroph.

Biological material for examination

CLASS . Chlorophyceae

CW Group: *Chlamydomonas, Gonium, Pandorina, Eudorina, Volvox*

DO Group: *Pediastrum, Hydrodictyon*

Oedogoniales: *Oedogonium*

CLASS . Trebouxiophyceae

Eremosphaera

Trebouxia

CLASS . Charophyceae

Order Charales: *Chara, Nitella*

Order Coleochaetales: *Coleochaete*

Order Zygnematales: *Spirogyra, Cosmarium, Staurastrum*

Order Klebsormidiales: *Klebsormidium*

Phylum EUGLENOPHYTA or Euglenoids

Euglena

Phacus

Trachelomonas

Procedure

Prepare microscope slides with the green algae as instructed. To observe pyrenoids add a drop of Lugol Solution (IKI). Always observe your samples at lower magnification first, then proceed at higher magnification.

1. In unicellular and colonial forms (Chlamys and other Volvocales) label the cell wall, plastid, pyrenoid, and flagella. Use the taxonomic key provided to identify the genera

2. In the cenobial *Pediastrum* and *Hydrodictyon* (the water-net) label cell wall, plastid, and pyrenoid

3. With the filamentous *Oedogonium*, label cell wall, plastid, pyrenoid, apical ring or cap, identify the female thallus and observe the oogonium and egg, look for the male filaments and observe antheridia

4. With the unicellular *Eremosphaera*, observe the cell wall and distribution of plastids

5. With the lichen sample provided make cross sections and prepare wet mounts. Look for the unicellular green cells, *Trebouxia*, label the cell wall and plastids, observe the colorless fungal filaments holding together the green cells

6. Desmids: Observe the characteristics of the cell: semicells and isthmus, and ornamentations on the cell wall. Use the taxonomic key provided to identify the genera

7. With *Spirogyra* slide observe the filaments with the spiral plastid, pyrenoids and nuclei. In the sexual reproduction slides label the conjugation tube, gametes, zygote, and determine the conjugation type (scalariform or lateral).

8. In *Klebsormidium*, observe the filament, cells, plastids and cell wall

9. In *Coleochate*, observe the general morphology of the thallus, cell wall, plastid and pyrenoid

10. *Chara* and *Nitella:*
    a. Place your specimen in a Petri dish and make observations of the macroscopic thallus using a dissecting scope. Label the rhizoid and the erect portion with nodes and internodes, apex, and whorl of branches

    b. Using a razor blade make cross sections of the thallus, prepare a wet mount, and observe central cell and cortical cells (if any)

    c. Transfer the reproductive structures to another slide and observe at the microscope. Identify globule and nucule

        i) The female nucule contains an egg surrounded by spiral cells and ending in a crown of cells at the apex

        ii) The male globule consists of several shield cells. On the wet mount press firmly with the pencil eraser to separate the shields and observe the released antheridial filaments

11. In Euglenoids observe the general morphology of the thallus and metaboly if present. Label pellicle, flagellum, chloroplast, muciferous bodies, reservoir, stigma, lorica, paramilon, and nucleus

Results

1. Name the genera found with the following morphologies:
    a. Unicellular forms:
    b. Colonial forms:
    c. Coccoid cenobial forms:
    d. Monadoid cenobial thalli:
    e. Filamentous thalli:
    f. Parenchymatic forms:
    g. Parietal plastids:
    h. Reticulated plastids:
    i. Central (Axial) plastids:
    j. Type of conjugation in *Spirogyra*:
    k. Metaboly:
    l. Rigid periplast:
    m. Flexible periplast:
    n. Lorica:
2. How can you distinguish between *Cosmarium* and *Staurastrum*?
3. How can you distinguish between *Chara* and *Nitella*?
4. How many spiral cells did you find in the Nucule?
5. What is the fate of the zygotes in Chlorophyceae, Trebouxiophyceae, and Charophyceae?

6. Compare the Charophycean orders using the following table

	Chlorokybales	Klebsormidiales	Zygnematales	Desmidiales	Coleochaetales	Charales
Cells						
Thallus						
Life cycle						

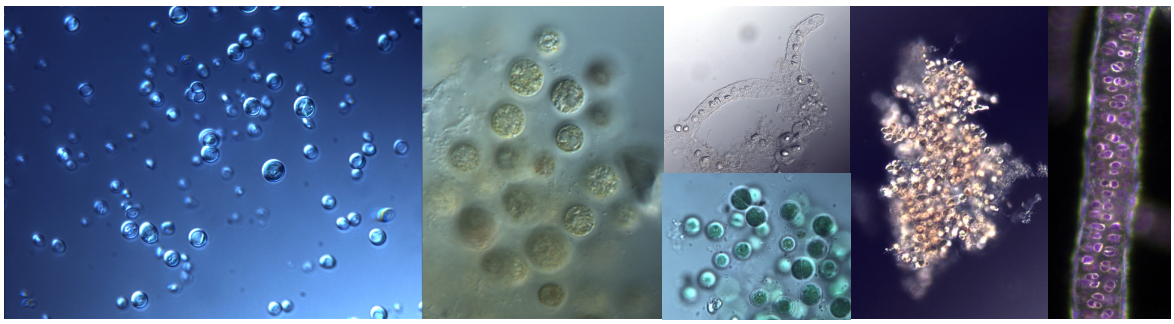
7. Compare the green algal classes with the following table:

	Prasinophytes	Ulvophyceae	Chlorophyceae	Trebouxiophyceae	Charophyceae
Cells (uni- or multinucleated)					
Cytokinesis (phycoplast or phragmoplast)					
Thallus (simple, branched, uniseriated, coenocytic)					
Life cycle (sporic, gametic, zygotic; iso- or heteromorphic)					
Flagella (iso- or heterokont; apical or lateral)					
Habitat (Freshwater, marine, or terrestrial)					

8. Euglenoids are green but they are not chlorophytes; what are the main differences between euglenoides and chlorophytes in terms of their photosynthetic pigments AND food storage?

Rhodophyta I

THE RED ALGAE: BASAL RHODOPHYTES



Examples of several types of basal rhodophytes: *Cyanidium, Rhodella, Stylonema, Porphyridium, Rufusiella and Bangia.*

Introduction

The Red algae are a diverse marine group with a wide range of morphologies. Although unicellular and colonial thalli are known, most rhodophytes are filamentous in nature with the pseudoparenchymatic thallus as the most common morphology. The cell wall contains a fibrous component made out of cellulose and a matrix component made out of complex gels or phycocolloids (agars and carrageenans). The red color is due to R-phycoerythrin, and chlorophyll *a* is the main green pigment. Floridean starch is the storage product. Flagella are absent in red algae.

Classification

PHYLUM RHODOPHYTA

I. CLASS CYANIDIOPHYCEAE: *Cyanidium*
II. CLASS RHODELLOPHYCEAE: *Glaucosphaera, Rhodella*
III. CLASS STYLONEMATOPHYCEAE: *Stylonema, Chroodactylon, Rufusia*
IV. CLASS PORPHYRIDIOPHYCEAE: *Porphyridium*,
V. CLASS COMPSOPOGONOPHYCEAE: *Compsopogon, Boldia*
VI. BANGIOPHYCEAE or Bangiophyceans: *Bangia, Porphyra*

VII. CLASS FLORIDEOPHYCEAE or Florideans
   a. SUBCLASS HILDENBRANDIOPHYCIDAE: *Hildenbrandia*
   b. SUBCLASS NEMALIOPHYCIDAE
      i. Order Order Acrochaetiales: *Acrochaetium*
      ii. Order Palmariales: *Palmaria, Halosaccion*
      iii. Order Batrachospermales: *Batrachospermum*
      iv. Order Corallinales: Coralline algae
      v. Order Nemaliales: *Nemalion*
   c. SUBLASS AHNFELTIOPHYCIDAE
      i. Order Ahnfeltiales *Ahnfeltia*
      ii. Order Pihiellales
   d. SUBCLASS RHODYMENIOPHYCIDAE
      i. Order Rhodymeniales: Rhodymenia
      ii. Order Gigartinales: *Chondrus, Gigartina*
      iii. Order Gracilariales: *Gracilaria*
      iv. Order Halymeniales: *Halymenia, Sebdenia*
      v. Order Nemastomatles: *Nemastoma, Schyzymenia*
      vi. Order Plocamiales: *Plocamium, Sarcodia*
      vii. Order Gelidiales: *Gelidium*
      viii. Order Ceramiales: *Polysiphonia*

Biological material for examination

CLASS PORPHYRIDIOPHYCEAE: *Porphyridium*, CLASS

COMPSOPOGONOPHYCEAE: *Boldia*

BANGIOPHYCEAE or Bangiophyceans: *Bangia, Porphyra*

Procedure

Prepare microscope slides with the red algae as instructed. Observe your samples at lower magnification first and then proceed to higher magnification.

1. In the unicellular form *Porphyridium* label cell wall, star-shape plastid, and pyrenoid
2. In the filamentous *Bangia*, label cell wall and plastid
3. In *Porphyra* observe the cell wall and plastids. Make cross sections and identify the layer (s) of cells in this species

Results

1. Name the genera with the following:
    a. Unicellular forms:
    b. Filamentous forms:
    c. Laminar forms:
    d. Monostromatic condition:
    e. Macrothallus:

2. How can you distinguish between *Bangia* and *Porphyra*?

3. How many layers of cells did you observed in *Porphyra*?

4. What is the function of the microthallus or cryptic phase *Conchocelis*?

Rhodophyta II

THE RED ALGAE: FLORIDEOPHYCEAE OR FLORIDEANS

Examples of red algae: Gracilaria, Polysiphonia, Bostrychia, Grateloupia and Champia

Introduction

This is the second part of the red algal survey. In this session we will be studying representatives of the Florideans.

Biological material for examination

CLASS FLORIDEOPHYCEAE or Florideans

SUBCLASS NEMALIOPHYCIDAE

Order Batrachospermales: *Batrachospermum*

Order Corallinales: Coralline algae

SUBCLASS RHODYMENIOPHYCIDAE

Order Rhodymeniales: *Rhodymenia*

Order Gigartinales: *Grateloupia*

Order Gracilariales: *Gracilaria*

Order Gelidiales: *Gelidium*

Order Ceramiales: *Polysiphonia, Callithamnion, Centroceras*

Procedure

Observe your samples at the dissecting scope. Prepare slides with the red algae as instructed, making thin cross sections when needed. Observe your samples at lower magnification first, then proceed at higher magnification. Add a drop of brilliant cresyl blue if necessary.

1. In *Batrachospermum* observe the general morphology of the thallus labeling the node and internode, axial cell, cortical cells, whorl branches, and pit connections. Look for evidence of reproductive structures at the tips of young branches.

2. In coralline algae observe at the dissecting scope both the crustose and articulated (geniculated) thalli. Label genicula and intergenicula. Look for evidence of reproductive structures in conceptacles.

3. In *Gracilaria* and *Grateloupia* observe the general morphology of thalli. Observe the apex and identify the apical cell(s). Make cross sections and observe the medulla. Identify cortical cells and determine type of medulla (cellular or filamentous).

4. In *Gelidium* observe the general morphology of the thallus. Identify the apical cell(s). In cross sections observe the medulla and identify the rhizines.

5. Prepare a microscope slide with *Callithamnion*. Observe the morphology of the thallus and the apical cell. Determine the branching type: is it alternate, opposite, or dichotomous? Label pit connections. Look for evidence of reproductive structures.

6. Prepare a wet mount with *Centroceras*. Observe the general morphology of the thallus and determine the branching type. Label cortical cells, spines, nodes, internodes, and forcipited apex.

7. *Polysiphonia*: place your sample in a Petri dish and observe at the dissecting scope. Make a wet mount to observe at the microscope the polysiphonous structure of the thallus, the apical cell(s), colorless uniseriate branches or trichoblasts, and pit connections. Then proceed to make cross sections of the axis and identify the central cell and determine the number of pericentral cells.

8. Request a prepared slide of *Polysiphonia* and observe at the microscope. Three different specimens are in the same slide. Proceed to identify each one of them as follows:

    a. Male Gametophyte: spermatangial branch, spermatangia, and spermatium

    b. Female Gametophyte: cystocarp, ostiole, carposporophyte, pericarp, and carpospores

    c. Tetrasporophyte: tetraspores, identifY the type of tetrasporangium (zonate, cruciate or tetrahedral)

Results

1. Name the genera found with the following morphologies:
    a. Uniaxial forms:
    b. Multiaxial forms:
    c. Filamentous medulla:
    d. Cellular medulla:
    e. Polysiphonous thallus:
    f. Rhizines:
    g. Conceptacles:

2. How can you distinguish between uniaxial and multiaxial forms?

3. From the studied samples which genera are agarophytes:

    And which ones are carrageenophytes:

4. What type of branching (alternate, opposite, dichotomous) did you observe in:

    Gelidium:

    Callithamnion:

    Centroceras:

    Polysiphonia:

5. What type of tetrasporangium (zonate, cruciate, or tetrahedral) did you observe in *Polysiponia*?

6. Identify the ploidy level (1N or 2N) for each structure in *Polysiphonia:*

Male gametophyte_____ Carposporophyte_____

Spermatangial branch_____ Pericarp_____

Spermatium_____ Carpospores_____

Female gametophyte_____ Tetrasporophyte_____

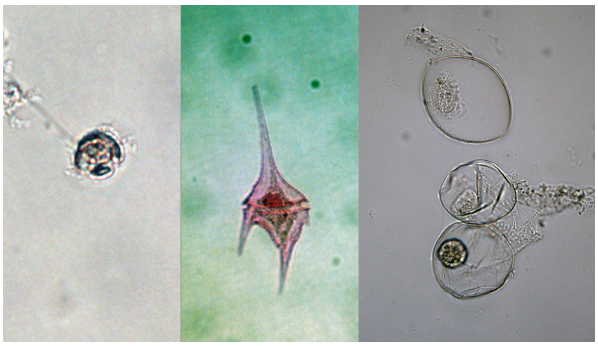
Cystocarp_____ Tetraspores_____

7. Compare the Rhodophytan classes using the following table:

	Example (genus)	**Thallus** (unicel, colonial, filament)	**Reproduction** (Asexual or Sexual)	**Life cycle** (Sporic, gametic, or zygotic; 1-, 2- or 3-phases; isomorphic or heteromorphic)
Cyanodiophyceae				
Rhodellophyceae				
Stylonematophyceae				
Porphyridiophyceae				
Compsopogonophyceae				
Bangiophyceae				
Florideophyceae				

Cryptophyta, Haptophyta & Dinophyta

THE CRYPTOMONADS, HAPTOPHYTES AND DINOFLAGELLATES



Coccolithophora (Haptophyta), *Ceratium and Pyrocystis* (Dinophyta)

Introduction

Cryptophytes are small flagellates very palatable to aquatic herbivores, their plastids contain chlorophyll *a* and *c*, as well as phycobilins (either phycoerythrin or phycocyanin). Storage is in the form of starch. Thallus is covered with a periplast made of proteins.

Haptophytes are mostly unicellular monadoid forms with chlorophylls *a* & *c*, and a yellow pigment, fucoxanthine. A reserve vacuole of chrysolaminarine is evident as well as the flagella and the haptonema. Thallus may be covered with organic or calcified scales (coccoliths).

Dinoflagellates are mostly unicellular, with chlorophylls *a* & *c*, and a brown pigment, peridinin. Granules of starch are the food storage. Two flagella, one smooth and one hairy, are present on two distinctive depressions of the cells, the cingulum and the sulcus. A cellulosic theca covers the cell below the plasmalemma.

Biological material for examination

PHYLUM CRYPTOPHYTA or Cryptomonads
Cryptomonas

PHYLUM HAPTOPHYTA or Coccolithophorids
Coccolithophora

PHYLUM DINOPHYTA or Dinoflagellates
Gymnodinium
Peridinium
Ceratium
Pyrocystis

Procedure

Prepare microscope slides with the algal samples as instructed. Observe your samples at lower magnification first, then proceed at higher magnification. Add a drop of brilliant cresyl blue or IKI (Lugol) solution when necessary.

1. Prepare a wet mount of *Cryptomonas* and label periplast, flagella, vestibulum and plastids.

2. In Coccolithophorids observe and label chloroplasts, coccoliths, flagella, haptonema, and chrysolaminarin vacuole

3. In Dinoflagellates observe and label epicone, hypocone, starch, nucleus, sulcus, cingulum, and flagella

    a. In naked forms observe chloroplasts

    b. In armored forms observe plates, apical and antapical horns

Results

1. Name the genera with the following morphologies:
    a. Naked dinoflagellates:
    b. Armored dinoflagellates:
    c. Coccoliths:

2. What is the difference between hypocones and epicones?

3. Coccolithoporids are gold in color, what is the pigment responsible for this?

4. Dinoflagellates are brownish, what pigment is responsible for this color?

5. What color was your *Cryptomonas* specimen? _____what pigment (s) are responsible for this coloration?

6. Compare the three algal groups with the following table

	Cryptophyta	Haptophyta	Dinophyta
Pigments			
Storage product			
Cell covering			

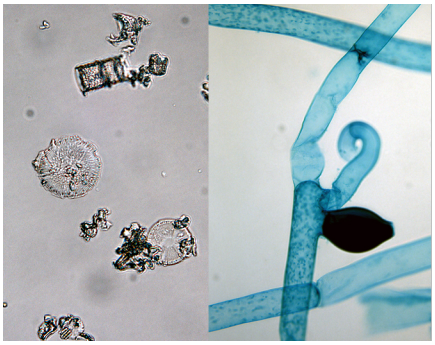
8. Take home and care of your new Dino pet. Do not expose to direct light but do not keep it in complete darkness, avoid shaking it, do not hold it with your hands too long (high temperature will kill it). If is well kept, it can last for 2-4 months. I keep mine by my bed table. Just after sunset tap it delicately or invert the tube gently to observe the blue glow of bioluminescence. This process will take place all night long and will stop before sunrise. This bioluminescence is an example of a circadian rhythm.

How some dinos such as *Pyrocystis* produce light?

How can you train your new Dino pet to light in the middle of the day?

Photosynthetic Stramenopiles I

DIATOMS TO YELLOW-GREEN ALGAE



Fossil diatoms and Vaucheria

Introduction

Stramenopiles algae are also called Heterokontophyta, Chromophyta, or Ochrophyta, they are characterized by chlorophylls *a* & *c*, and heterokont flagella with mastigonemes. Together with the Oomycetes they form the Stremanopiles group. Reserves are vacuoles of chrysolaminarin and lipids. The Stramenopiles is a diverse group of algae ranging from unicellular to massive forms such as giant kelps. Several classes are grouped together in this phylum including the famous diatoms and others. The brown algae or Phaeophyceae, also a stramenopile group, will be discussed next week. Classification of Stramenopiles according to your textbook is as follows:

CLASS	COMMON NAME
Bacillariophyceae	Diatoms
Bolidophyceae	Bolidophytes
Raphidophyceae	Raphidophytes
Chrysophyceae	Chrysophytes/Golden algae
Synurophyceae	Synurophytes/Golden algae
Eustigmatophyceae	Eustigmatophytes
Dictyochophyceae	Dictyochophytes
Pelagophyceae	Pelagophyceae

Pinguiophyceae	Pinguiophytes
Phaeothamniophyceae	Phaeothamniophytes
Chrysomerophyceae	Chhrysomerids
Xantophyceae/tribophyceae	Xantophytes/Tribophytes/Yellow-green algae
Phaeophyceae	Phaeophytes/Brown algae

Biological material for examination

CLASS BACILLARIOPHYCEAE or diatoms

Living samples (marine and freshwater)

Fossil diatoms (Diatomite or Diatomaceous earth)

CLASS CHRYSOPHYCEAE or golden algae

Ochromonas sp.

CLASS SYNUROPHYCEAE or golden algae

Synura sp.

CLASS TRIBOPHYCEAE or yellow green algae

Botrydium sp.

Tribonema sp.

Vaucheria sp.

Procedure

Prepare microscope slides with the algal samples as instructed. Observe your samples at lower magnification first then proceed at higher magnification. Add a drop of brilliant cresyl blue or IKI (Lugol) solution when necessary.

1. In Diatoms (living and fossils) observe the general morphology of the thallus, labeling frustule, epitheca, hypotheca, epivalve, hypovalve, valvar view, girdle view, chloroplasts and lipids (if any), and markings on the frustule

2. In *Ochromonas* note cell shape, chloroplast, flagella, and chrysolaminarin vacuole

3. In colonial *Synura* look for silica scales, chloroplast, flagella, and chrysolaminarin vacuole

4. In *Botrydium* observe the unicellular nature of the thallus and distribution of chloroplasts

5. In *Tribonema*, examine the filamentous nature of the thallus, distribution of chloroplasts and H-pieces of the cell wall

6. With fresh samples of *Vaucheria* notice the tubular thallus, chloroplasts, central vacuole and nuclei

7. In the *Vaucheria* prepared slide: observe the reproductive structures: antheridium and oogonium

Results

1. Name the genera found with the following morphologies:
    a. Coccoid thalli
    b. Unicell monadoid
    c. Colonial monadoid
    d. Coenocytic
    e. Filamentous
    f. Silica scales
    g. H-pieces
    h. Antheridium & oogonium

2. What is the difference between epitheca and hypotheca?

3. *Ochromonas* and *Synura* are golden algae, what is the main pigment responsible for their color?

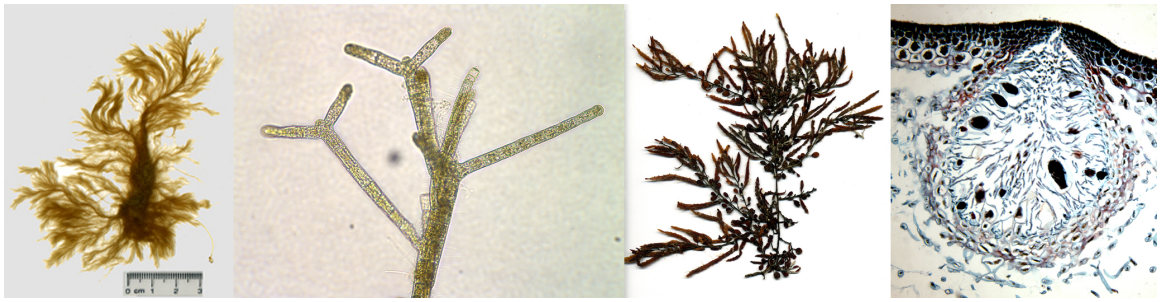
4. *Botrydium, Tribonema* and *Vaucheria* are yellow-green in color, what pigments are responsible for this coloration?

5. What type of sexual reproduction did you observe in *Vaucheria*?

6. Tribophyceae (a stramenopile) and Chlorophyceae (a green alga) are similar in color and morphology (parallel evolution), how can you differentiate both groups using a simple lab technique?

Photosynthetic Stramenopiles II

THE BROWN ALGAE OR PHAEOPHYCEAE



Examples of marine brown algae: *Ectocarpus, Sphacelaria, Sargassum and Fucus*

Introduction

The brown algae or Phaeophyceae are seaweeds with multicellular thalli, some of them forming extensive submarine forests. Their pigmentation is brown because of the excess of fucoxanthine in addition to the chlorophylls *a* & *c*. The food reserve is chrysolaminarine and mannitol. The cell wall, made of cellulose, contains an economically important gel known as alginic acid or alginates.

Biological material for examination

CLASS PHAEOPHYCEAE

ORDER Dictyotales

Family Dictyotaceae: *Dictyota*! !

ORDER Sphacelariales

Family Sphacelariaceae: *Sphacelaria*

ORDER Ectocarpales

Family Ectocarpaceae: *Ectocarpus*

Family Scytosiphonaceae: *Scytosiphon*

ORDER Laminariales

Family Laminariaceae: *Laminaria*

ORDER Fucales

Family Fucaceae: *Fucus*

Family Sargassaceae: *Sargassum*

Procedure

Prepare microscope slides with the algal samples as instructed. Observe your samples at lower magnification first, then proceed at higher magnification. Add a drop of brilliant cresyl blue stain when necessary.

1. In *Ectocarpus* observe the general morphology, label cell wall, chloroplasts, and physodes, determine branching type and presence of unilocs or plurilocs

2. In *Sphacelaria* notice the shape of the thallus, labeling the apical cell, uniseriate and multiseriate filaments. Observe the asexual tri-radiate propagules

3. In *Dictyota* observe the type of branching. Locate the apex and determine number of apical cell(s). Identify medullar and cortical cells in a cross section

4. In *Scytosiphon* observe the cylindrical thallus and constrictions

5. In *Laminaria* examine the morphology of the kelp, label holdfast, hapteria, stipe, intercalary meristem, and blade.

6. In the prepared slide of *Laminaria* observe the medulla, cortical cell, reproductive structures in sori, and identify paraphyses and sporangia

7. In *Fucus* note the laminar thallus, branching pattern, central vein, pneumatocysts, receptacles and conceptacles.

8. In the prepared slide of *Fucus* observe the reproductive structures and determine their sexual status:
    a. Male Gametophytes: male conceptacles, spermatangial branches, spermatangia, ostiole, and paraphyses
    b. Female gametophytes: female conceptacles, oogonial branches, oogonium, egg, paraphyses, and ostiole

9. In *Sargassum* notice the general morphology of the thallus: label holdfast, stipe, blades, and pneumatocysts

Results

1. Name the genera found with the following morphologies:
    a. Filamentous thalli
    b. Tubular thalli
    c. Laminar thalli
    d. Complex (with root-like, stem-like and leaf-like) thalli

2. Type of growth (diffuse, apical, intercalary, meristoderm) as observed in:
    a. *Ectocarpus*
    b. *Sphacelaria*
    c. *Dictyota*
    d. *Laminaria*
    e. *Fucus*

3. Name the genera found with the following life cycles types:
    a. Isomorphic
    b. Heteromorphic
    c. Monophasic

4. What is the function of a propagule and where did you find it?

5. What is the function of paraphyses and where did you find them?

6. Compare the Phaeophycean orders according to the following table:

	Dictyotales	Sphacelariales	Ectocarpales	Laminariales	Fucales
Life cycle					
Growth mode and Meristems					
Sexual reproduction					

7. Construct a table to compare the most important Stramenopiles classes:

	Pigments	Food storage	Thallus type
Bacillariophyceae			
Chrysophyceae			
Synurophyceae			
Xanthophyceae			
Eustigmatophyceae			
Phaeophyceae			

Algal Collection Guidelines

Collecting and preserving seaweeds at Dauphin Island Sea Lab, AL

Introduction

The habitats were algae can be found in the Southeastern USA are diverse, from the bottom of the photic zone of the Gulf of Mexico to the canopy of forest trees, inside or outside of animals (endozoic or epizoic algae, respectively) and plants (endophytic or epiphytic algae, respectively), and even in symbiosis with fungi (lichens), plants (aquatic fern *Azolla*), or animals (hydras, corals). However, it is the aquatic habitat in which wer find the higher diversity and abundance of algae. Floating masses of green algae can be found on the surface of lakes or ponds (genera such as *Spirogyra*, *Zygnema, Mougeotia, Rhizoclonium* and others are common). Attached to the bottom (benthic algae) of these freshwater habitats we can found larger characeans (*Chara* and *Nitella*). Microscopic algae can be found attached to submersed vegetation as epiphytes (such as *Ulothrix, Oedogoniu*m, *Chaetophora*, and many others). In the plankton of these freshwater habitats it is common to find unicellular and cyanobacteria (*Anabaena, Merismopedia*), chlorophytans (*Scenedesmus and Pediastrum*), and diatoms such as *Gyrosigma, Fragillaria*, and many others.

Estuarine habitats, with a constant exposure to a salinity gradient, provide an excellent area to observe macroalgal genera such as *Bostrychia, Caloglossa, Vaucheria, Ulva* and *Entermorpha*. In marine environments, seaweeds form notable populations. In the intertidal zone, subjected to constant immersion and emersion by tides, it is common to observe *Ulva, Entermorpha, Gelidium, Ectocarpus, Cladophora*, and *Chaetomorpha*, among others. In the subtidal zone seaweeds such as *Gracilaria, Grateloupia, Digenea, Spyridia,* and *Polysiphonia*, are common. These seaweeds in turn are used as living substrates by other microscopic epiphytic algae of the genera *Entocladia, Ulvella, Giffordia, Goniotrichum, Erythrotrichia, Acrochaetium* and many diatoms such as *Navicula* and *Grammatophora*. Representatives of the marine phytoplankton

are also diverse and include diatoms and dinoflagellates. Floating masses of seaweeds or sargasso are also common in the Gulf. These pelagic algae are basically brown algae from the genus *Sargassum*.

Algae are not strictly aquatic organisms. Some can be found on terrestrial habitats as well, living on soils (edaphic algae) or above the soil (subaerial algae). Terrestrial and subaerial habitats are populated with green and blue-green algae in amounts easily detected by the naked eye. Subaerial algae are usually found living on rocks (epilithic algae), bark (epiphellemic algae), and leaves (epiphyllous or foliicolous algae) especially when atmospheric humidity is relatively high.

Collection

Benthic freshwater algae are collected by hand or with the aid of scrappers or knives. Epiphytic algae are collected along with their plant substrate. Delicate algae are placed in small vials and larger algae in plastic bags.

Benthic marine algae are collected in a similar fashion, but it is important to obtain the base of the alga with the aid of a scrapper. Place your collection of seaweeds in a bucket or large bag, then transfer individuals to smaller vials or bags.

Planktonic algae or phytoplankton will be collected with the aid of a special net with a conical shape and containing a collecting bottle at the end. The concentrated sample will be transferred to small vials.

Terrestrial algae will be collected by hand with the aid of scrappers or knives and placed in plastic bags.

Labeling

All plastic bags and vials should be individually labeled. Use permanent markers to label your plastic bags. Write down all the information obtained in field into your field notebook using pencils or permanent markers. Data that you must include for each and every one of your sample bags or vials are:

- Locality: Place, County, State, if possible use a GPS for exact location
- Date of collection
- Collector (this is probably your name or somebody else)
- Habitat and Observations:
  - ✓ Substrate where the alga was found (rocks, epiphytic, on sand, etc).
  - ✓ If intertidal, above or below the intertidal, etc.
  - ✓ Color, consistency, abundance, etc.

Preservation

For morphological studies, the algae are preserved in a 5% solution of formaldehyde or 70% Ethanol. The solution should be prepared with water from the same location, for example, freshwater samples should be fixed using fresh water, and marine algae should be fixed using seawater. All material should be protected from direct sunlight inside a container such as a bucket or ice chest or a dark bag.

Subaerial algae can be air dried in the shade and maintained in a bag or vial away from direct sunlight.

For DNA analysis, algae are preserved using a desiccant, silica gel crystals. The alga or (in case of larger algae) portions of the alga are placed in Ziploc bags with enough silica gel. Ethanol (70%) can also be used for this purpose.

In cases where formaldehyde or silica gel is not at hand, I follow this procedure: I let the algae to air dry for a while (out of direct sunlight). After one hour or so, I place the algae on newspapers and I proceed to roll them up (like sushi). I found this method to work fine with marine algae. After arrival to the lab all samples are re-wet as soon as possible for a couple of hours in seawater, then sorted, and fixed with formaldehyde or ethanol as described above.

Herbarium

In order to make a herbarium specimen with your alga, follow this protocol if your sample is macroscopic (seaweeds or larger freshwater algae):

a) Place the sample in a plastic tray with enough water (seawater or freshwater according to the source of the specimen) and clean the alga from sand or mud using forceps, needles, or a small brush
b) Label an adequate piece of herbarium paper (see Labeling above)
c) Place the herbarium paper in the water below the alga
d) Pull the paper with the alga out of the water
e) Use forceps or a brush to arrange the alga on the paper
f) Cover the alga with muslin
g) Place the sample in between blotter papers or newspapers
h) Place newspapers in between corrugated ventilators
i) Place your set in a plant press and close tight with straps
j) Change blotter papers/newspapers every day until sample is completely dried

In case of microscopic algae (phytoplankton or small epiphytes), prepare a semi-permanent slide following this protocol: make sure your sample is fixed first: 70% Ethanol or 3-5 % formalin, for 15-30 min for microscopic to delicate forms, and up to 24 hrs for thick, gelatinous or more solid algae such as seaweeds. After fixation rinse the specimen with distilled water. The Glycerin-Gel method for microscopic forms is as follows:

a) After fixation, stain with 1% aniline blue or lugol
b) Transfer to slide

c) Add warm glycerin-jelly (5 gr gelatin, 30 ml distilled water, 35 ml glycerin)
d) Cover with a coverslip
e) Store flat and face-up
f) Label microscope slide with permanent marker: indicate the position of the algae (circle) and write down all pertinent information (see Labeling section above)

Identification

The taxonomic determination of the algal name requires previous knowledge of its biology, particularly its morphology. With properly collected, labeled and preserved algae it is possible to identify the genus or in some cases the specific epithet (species' name). We will also need some specialized references (algal guides) and become familiar with the use of taxonomic keys. Sometimes it will be necessary to use special techniques to observe particular features of the algal morphology, such as pericentral cells, pyrenoids, medulla arrangement, coralline algae, etc. During laboratory sessions follow the instructions in order to use these taxonomic keys and how to proceed with special techniques.

Further information

Visit our website www.phycolab.ua.edu for the Alabama Seaweeds Database and other useful links.

The Smithsonian has a great website with useful information for collection and preservation of marine algae at http://www.nmnh.si.edu/botany/projects/algae/

Fieldtrip Report I

CAMPUS, THE UNIVERSITY OF ALABAMA, TUSCALOOSA, AL

Cyanobacteria	Chlorophyta	Rhodophyta	Others

Fieldtrip Report II

LAKE LURLEEN STATE PARK, TUSCALOOSA, AL

Cyanobacteria	Chlorophyta	Rhodophyta	Others

Fieldtrip Report III

DAUPHIN ISLAND SEA LAB, DAUPHIN ISLAND, AL

Cyanobacteria	Chlorophyta	Rhodophyta	Others

Algal Collection

FINAL REPORT

Your taxonomic list MUST corresponds to your algal collection order

Freshwater	Marine	Terrestrial
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20	1 2 3 4 5 6 7 8 9 10	1 2 3 4 5
Total FW:	Total Marine:	Total Terrestrial:

GRAND TOTAL: _____ algae