Biology 2015 – Evolution and Diversity Lab

Lab 4: Seedless Plants

Introduction

Last week you studied algae, the photosynthetic protists that include the green algal Charophytes that are the sister group to the land plants. You will study land plants both this week and next.

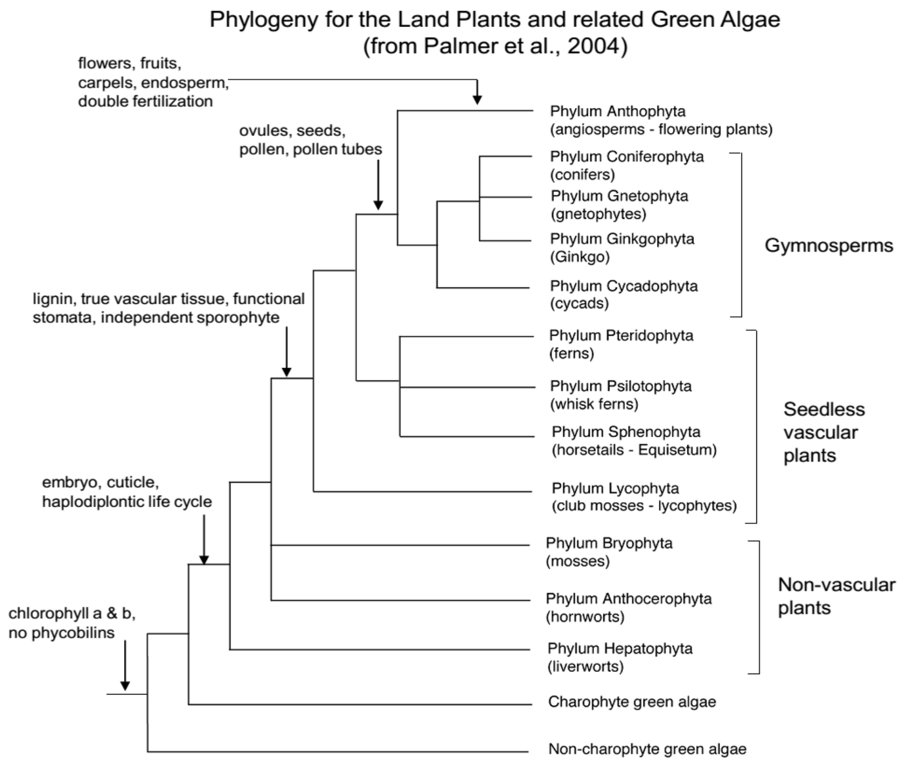

Figure 1. Phylogeny of land plants and related green algae.

Plants are a fascinating group as we will be able to observe many of the adaptations to living on land that occurred throughout their evolutionary history. The plants are distinguished from the green algae by three characteristics: the cuticle, the diploid embryo, and alternation of generations. The **cuticle** is a waxy, waterproof coating found on the aerial parts of a plant, which minimizes the plant's loss of water to the surrounding air. The cuticle is not found on the below-ground parts of a plant because it would inhibit the uptake of water and nutrients. For the same reasons, cuticles are not found in the algae.

The **embryo** is a developing, multicellular 'baby' plant that is nurtured and protected by the parent plant. The evolution of the cuticle and of the embryo facilitated the colonization of land by plants.

The seedless plants reproduce both sexually and asexually, but, as the name suggests, these plants do not produce seeds. Though seeds provide important advantages in both dispersal and establishment of offspring, seedless plants continue to be common in a variety of humid terrestrial environments.

The **haplodiplontic life cycle** is characteristic of plants. This life cycle may also be called **"diplohaplontic",** or be referred to as **"alternation of generations".** In a haplodiplontic life cycle, both multicellular haploid individuals and multicellular diploid individuals are produced. There are several examples of haplodiplontic life cycles in this handout.

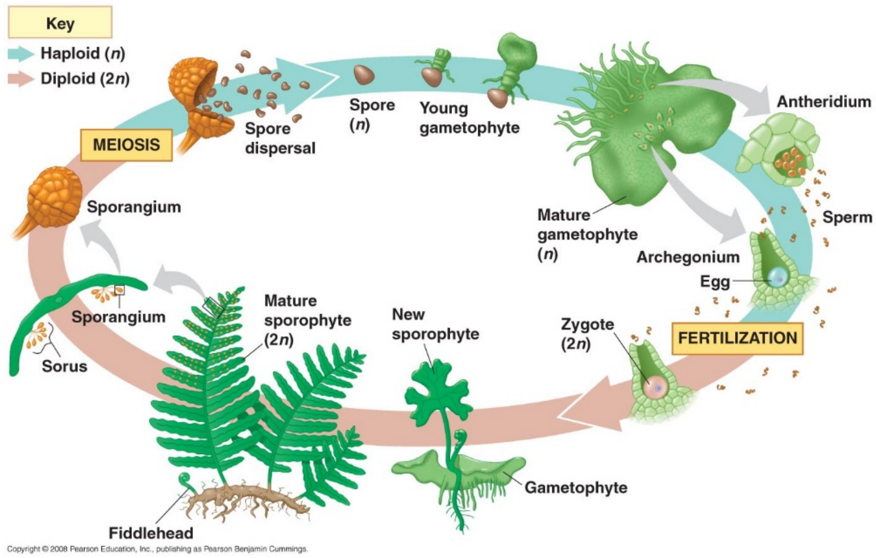

Figure 2. Haplodiplontic life cycle of ferns.

It will help to further introduce some new terms, as we will be using several new ones in the two coming labs. A "**gametophyte**" is the haploid multicellular organism that produces **gametes** (through mitosis); two gametes fuse to produce a **zygote.** The zygote, instead of going through meiosis, as in Charophite algae, forms a multicellular "**embryo**". The embryo later develops into the "sporophyte", which is the diploid multicellular organism that produces **spores** (through meiosis). Spores are haploid and they don't fuse with other cells; rather, they develop into a multicellular gametophytes.

It is convenient to consider 'seedless plants' as a group distinguishable from the seed plants, but *the seedless plants do not form a monophyletic group*. And though it's also convenient to divide the seedless plants into two subgroups based on the presence or absence of vascular tissues, these subgroups are not monophyletic either. The first subgroup of the seedless plants consists of the non-vascular plants, or "bryophytes". There are three clades within the bryophytes: Hepatophyta (the liverworts); Bryophyta (the mosses); and Anthocerophyta (the hornworts). You'll see examples of each clade in today's lab. There are about 24,000 known species of bryophytes. They are all small plants that mostly lack specialized vascular tissues. They have "dominant" gametophytes and "dependent" sporophytes.

The second subgroup contains the seedless vascular plants. Vascular plants produce two important tissues: **xylem** transports water and solutes throughout the plant and provides structural support; and **phloem** transports organic compounds. These tissues allow the vascular plants to grow much larger than the non-vascular bryophytes.

The seedless vascular plants include two phyla: Lycophyta (the "club mosses") and Pteridophyta (the ferns). You'll see examples of both in today's lab. Together these phyla include about 14,000 species, and the great majority of them are ferns. Seedless vascular plants were dominant during the Carboniferous, and much of our fossil fuel comes from their remains.

From an evolutionary perspective, the dominance of the gametophyte and the sporophyte have swapped in importance, going from a dominant gametophyte and dependent sporophyte, to a dominant sporophyte and a reduced and dependent gametophyte. As you will see in this and the following week, the gametophyte is the dominant stage in the “bryophytes”, while the sporophyte is short-lived and dependent on the gametophyte for nutrition. In vascular plants there’s been an important transition: the sporophyte is large and long-lived and, by contrast, gametophytes are small and short-lived.

MARCHANTIOPHYTA (Liverworts)

There are several living liverworts for you to study today. They include examples from the genera ***Marchantia.*** In Figure 3 we have the life-cycle of liverworts.

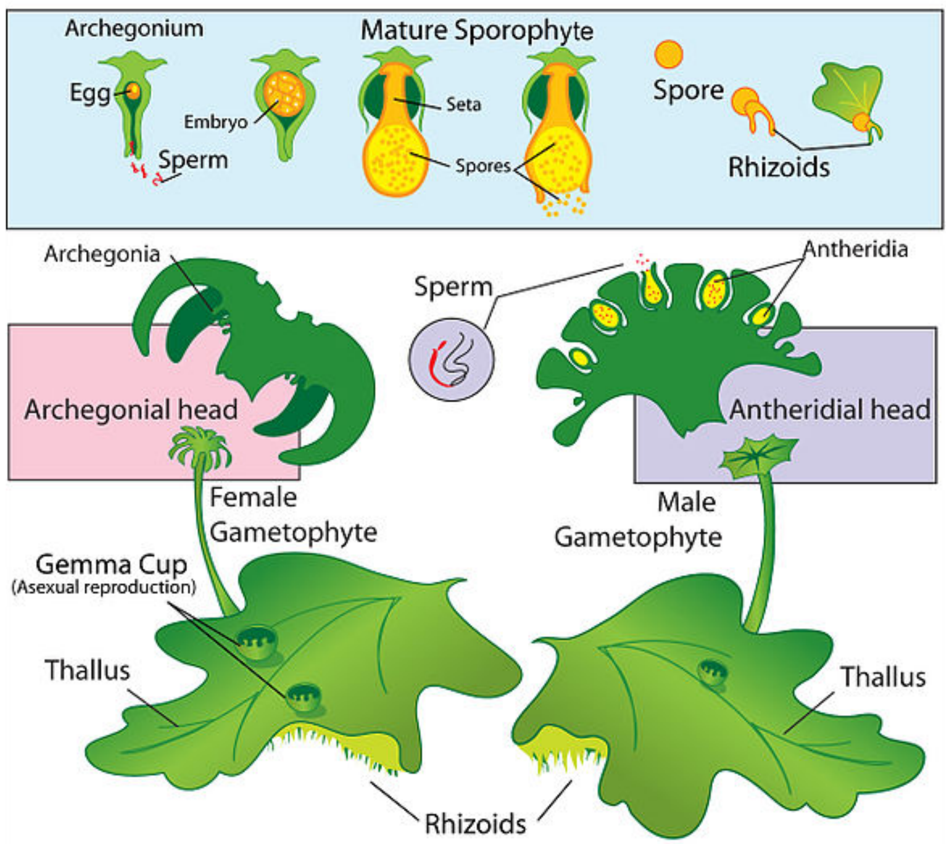

Figure 3. Liverwort life-cycle.

Liverwort Gametophyte Structure

There are two major liverwort groupings based on gametophyte morphology. The *Marchantia* specimens we have today are good examples of "thalloid" gametophytes (Figure 4). Thalloid plants are essentially flat layers of tissue.

But there are also "leafy" liverwort gametophytes that appear to have a "stem and leaves" morphology. You should examine *Bazzania* for an example of this form. Liverworts always grow in very moist places.

Examine the *Marchantia* specimens, and plan to use a magnifying glass for observing the smaller structures.

Use a magnifying glass to look for openings on the surface of the *Marchantia* thallus. Remember that the aerial portions of land plants are covered with cuticle, and that openings through the cuticle are needed for gas exchange.

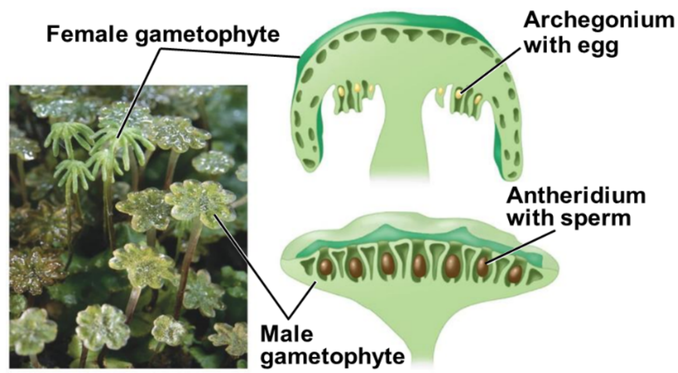

Figure 4. Examples of "thalloid" liverworts of the genus *Marchantia*.

- ***Find and examine prepared slides labeled "*****Marchantia*** ***Archegoniophore l.s." and "*****Marchantia** ***Antheridiophore l.s."***

Be sure to see the Archegonia and Antheridia on these slides (Figure 5). Knowing that liverworts live in wet, rainy places, and that liverwort sperm are motile, how do you suppose the structures of the antheridiophores and archegoniophores function in fertilization of the egg cells?

Figure 5. Archegonia (left) and antheridia (right) of *Marchantia*.

You should be able to find the structures called "**gemmae cups**" on the living specimens. Gemmae cups contain asexual propagules called "**gemmae**" (singular, "gemma"). When raindrops hit one of the cups, the gemmae can be dispersed in the splash. The gemmae are then able to grow into new gametophyte thalli. On the underside of the thallus there are root-like rhizoids. You may be able to see them if you gently lift an edge of the thallus.

- ***Find a prepared slide labeled "*****Marchantia** ***Gemma Cup. Sec.". Observe the structure of the gemmae cups and of the gemmae found inside.***

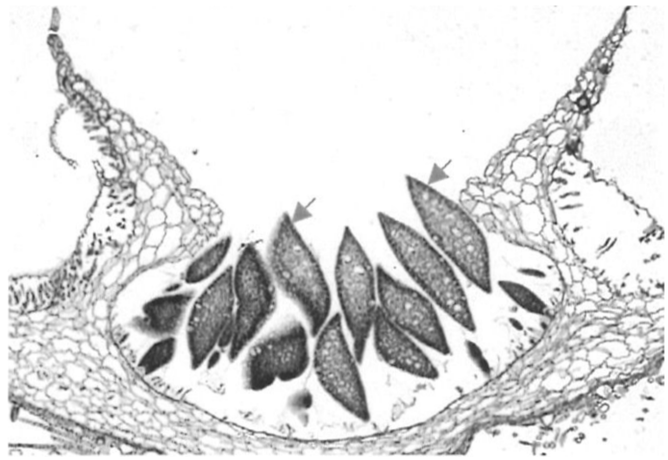

Figure 6: Gemmae cup. The gemmae (arrows) are roughly disk-shaped in face view.

Liverwort Sporophyte Structure

Liverwort sporophytes are quite small. They form within the archegonia on the underside of the archegoniophores. They consist of a relatively large sporangium (or "capsule"), a fairly short, stout seta, and a small "foot" as shown in the life cycle diagram of Fig. 10.

- ***Find a prepared slide labeled "Marchantia Sporophyte l.s." Find the sporophytes, and compare them to the one shown in Figure 7.***

How do the *Marchantia* sporophyte compare to the sporophyte of the bryophytes?

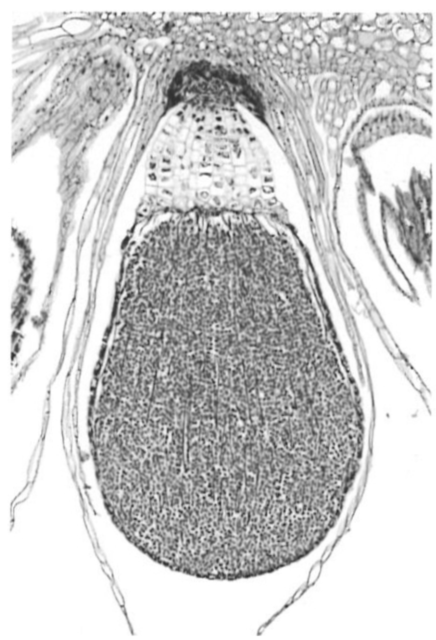

Figure 7. Sporophyte of *Marchantia*.

ANTHOCEROTOPHYTA (Hornworts)

For completeness, we'd like to show you an example of a hornwort but we have had difficulty growing hornworts in our greenhouse- they tend to dry out very easily. Fortunately we have prepared slides that show important structures.

Figure 8. A picture of a hornwort, with sporophytes (light green stalks) growing from the gametophytes.

Find and examine prepared slides labeled "Anthoceros, ."

BRYOPHYTA (Mosses)

We have several species of living moss for you to examine today, including examples from the genera ***Hypnum, Mnium, Dicranum, Polytrichum,*** and ***Sphagnum.*** We also have living moss examples that were collected "wild" in the Department greenhouses. And, in addition to the living specimens, we have slide preparations of *Polytrichum* and *Mnium.*

You should use a magnifying glass to observe the living, 'wild', moss examples to be sure you recognize the gametophytes and sporophytes, and can relate them to the life cycle diagram. Please treat the specimens gently.

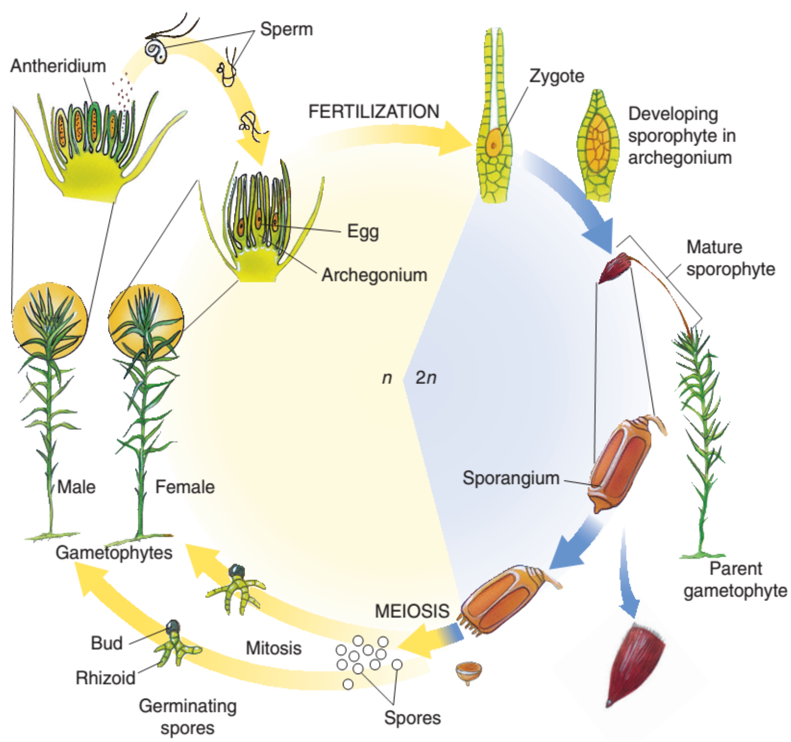

Figure 9. Life-cycle of a typical moss (Bryophyta).

Moss Gametophyte Structure

You'll notice that the gametophytes are green, and appear to have a "stem and leaves" structure - like what you'd expect of a plant. But true leaves and true stems are absent in non-vascular plants. And most mosses truly are "non-vascular". A few, however, and *Polytrichum* is one, do have xylem-like and phloem-like tissues in their "stems". These moss tissues likely arose independently of the true xylem and phloem found in the vascular plants, so this is an example of convergent evolution.

The gametophytes also have structures called **"rhizoids".** These are unicellular, filamentous structures that serve to anchor the gametophyte to the soil, and that also function in water and nutrient uptake.

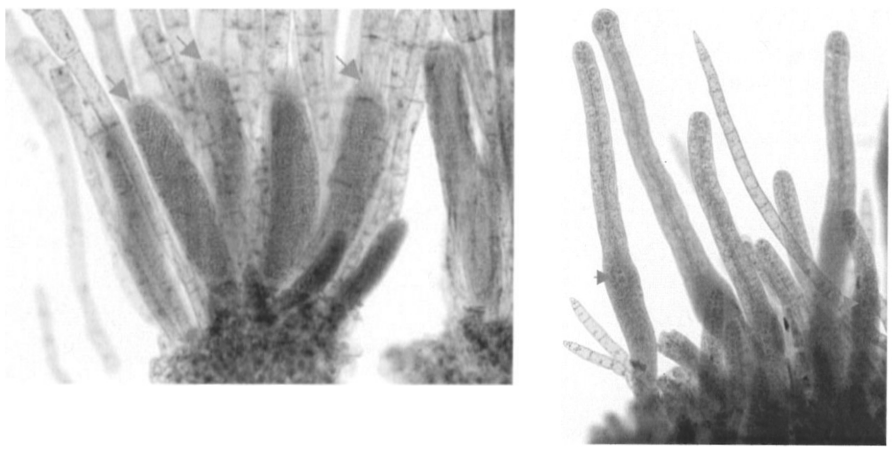

Figure 10. Moss antheridia (left, arrows) and archegonia (right). Look carefully at the thicker portions of the archegonia, indicated by arrows. Yo may be able to see the egg cells that are located there.

- ***Find a prepared slide labeled "Moss Antheridia and Archegonia w.m."***

Most mosses have separate male and female gametophytes. The gametangia are found clustered near the apex of their stalks. You probably won't observe gametangia on the living moss specimens, but the prepared slides will show the gametangia well.

The sperm released from the antheridia must reach an archegonium, and then swim down the neck canal to reach the egg cell. So water is necessary for sexual reproduction to occur in the mosses (and for other seedless plants). Rainfall may provide a continuous film of water for the sperm cells to swim in, but the splash of raindrops probably also helps many sperm cells to reach the female gametangia.

Moss Sporophyte Structure

- ***Examine the living mosses, with sporophytes, on display.***
- ***Examine the dried/preserved examples of Polytrichum sporophytes on display.***
- ***Find a prepared slide labeled "Mnium: capsule, median l.s."***

The sporophytes of the mosses consist mainly of a "capsule" supported and elevated by a stalk called a "seta". You've probably already noticed that the sporophyte grows right out of the archegonium in which it began development as an embryo. The capsules are the sporangia in which meiosis occurs and the spores form. Capsules that are green are immature; brown capsules may be shedding spores, or may already be empty. You should examine the prepared slides of *Mnium* capsules, and the dried/preserved sporophytes of *Polytrichum* that we have available.

Asexual Reproduction

- ***Find a prepared slide labeled "Moss protonema: with bulbils, w.m."***

Many plants are capable of asexual reproduction, and some of them produce structures that have this specific function. When moss spores germinate, a loose filamentous mass of cells called a "protonema" forms before development of an organized gametophyte begins. In some species, small globular structures called "bulbils" may form on short stalks that branch from the filaments of the protonema. These bulbils function as vegetative propagules. You should observe the prepared slides of moss protonemata that have produced bulbils.

Figure 11. Moss bulbils.

TRACHEOPHYTA (Ferns)

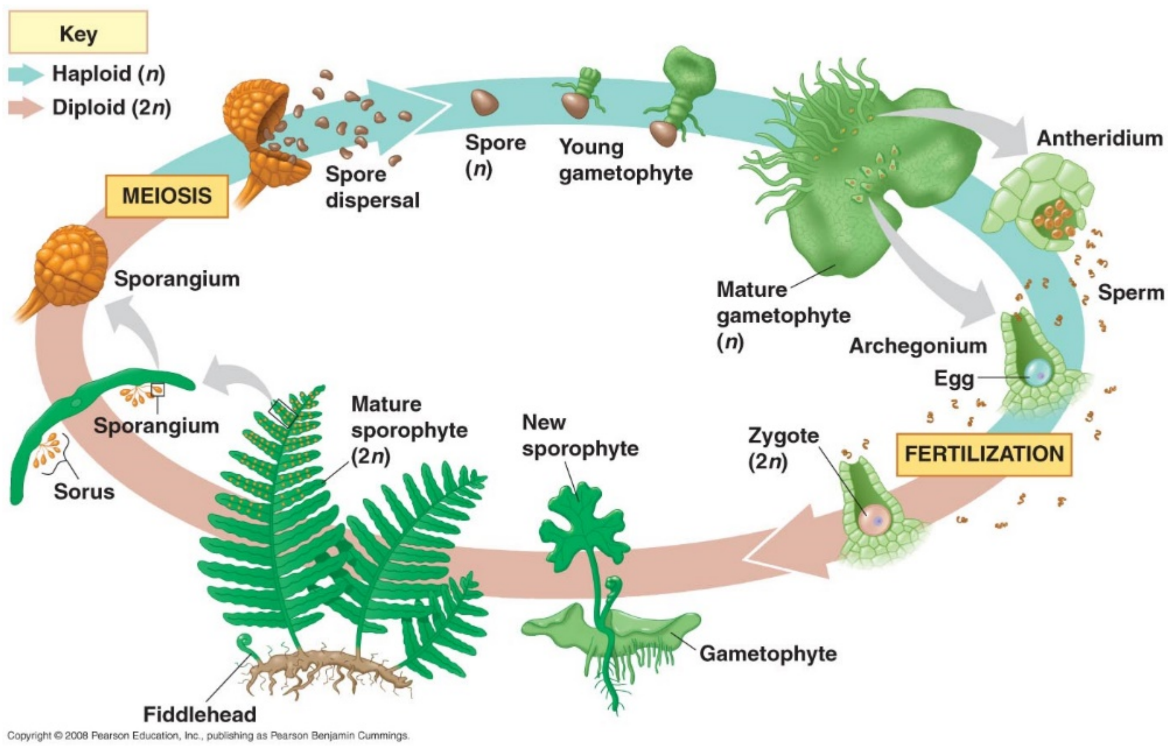

Figure 12. Life cycle of ferns (Pterydophyta).

We will start by describing the "typical" ferns, then cover the horsetails. Ferns are the most diverse of the seedless vascular plants. We have some of that diversity on display in the lab today.

Ferns do not have a big root system. Instead, they have a shallow **rhizoidal** system through which they obtain the materials they require for photosynthesis. Some fern genera, like *Azolla*, can even live on the surface of water. This is one of the ferns on display today. *Azolla* forms a mutualistic relationship with an organism you have already learned. Remove a leaflet, crush it with a razor blade, and mix it with a drop or two of water. What is the name of the symbiont?

Fern Sporophyte Structure

Ferns often have very large leaves- they are **megaphylls** that have multiple vascular strands. The leaves are often "compound" leaves, in that they are divided into several (or many) leaflets. In many ferns, there is no above ground stem; the leaves arise from underground **rhizomes,** from which the roots also branch.

- ***Find a prepared slide labeled "Fern Sporangia Mature c.s."***

In many ferns, including almost all of those we have on display, sporangia are borne on the undersides of the leaves, generally arranged in circular or elongated clusters (called **"sori",** singular **"sorus")** that are often tan to dark brown in color.

In many cases there will be a layer of tissue that partly covers the cluster of sporangia. It's called an **"indusium".** Examine the prepared slides of fern sporangia, and determine if an indusium is present.

Examine the various ferns on display. See if you can determine whether any sori are present, and if so, whether they are covered with an indusium.

- ***Find the fern leaf with sori on display at a dissection scope.*** Examine the sari. Is an indusium present? Examine the sporangia.

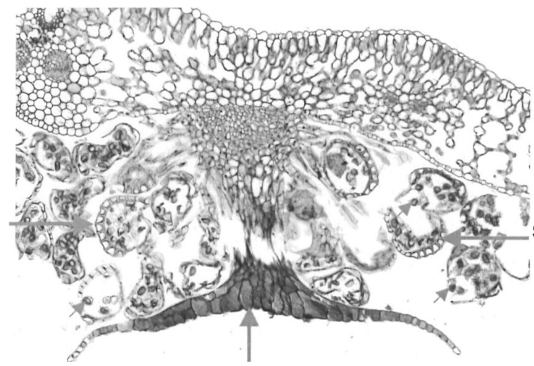

Figure 13. Section through a sorus and its indusium. Sporangia showing annular cells are labeled, and several sporangia contain spores (small arrows).

On the 'spine' of each sporangium is a row of thick-walled **"annular" cells** that contract as they dry out. This contraction causes the sporangium to split open, and the top of the sporangium - containing the spores - pulls back, and then suddenly snaps forward again. This movement 'catapults' the spores into the air. If you dry out some sporangia under the light of the ***dissection scope***, you may be able to witness this dispersal action.

Fern Gametophyte Structure

- ***Find a prepared slide labeled "Fern: young prothallia w.m."***

When the spores germinate, new gametophytes (also called **"prothallia")** are formed. Examine the prepared slides of young fern prothallia. You should be able to observe examples of germinated spores and the small gametophytes that have begun to develop. Notice that even the smallest of these new gametophytes has at least one rhizoid.

- ***Find a prepared slide labeled "Fern prothallium: antheridia*** & ***archegonia, w.m."***
- ***Observe the living fern prothallia (gametophytes) on display.***

Mature fern gametophytes are small, often heart-shaped, haploid plants. On their undersides there are rhizoids, and also antheridia and archegonia. You should examine the prepared slides of these prothallia. You should observe archegonia clustered near the 'cleft' of the heart-shaped thallus. You should

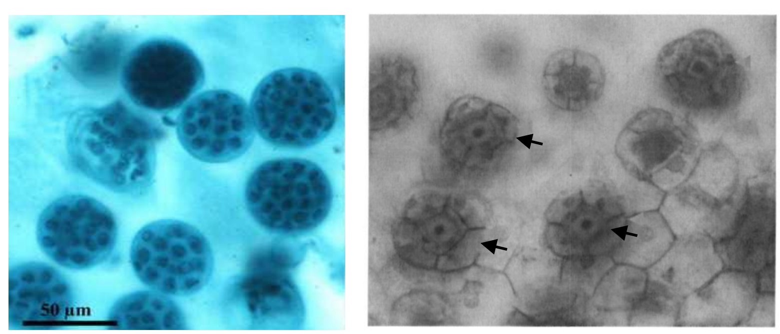

Figure 14. Fern antheridia (left) and archegonia (right). The 'best' of the archegonia are labeled with arrows.

observe antheridia near the clustered archegonia, and especially near the rhizoids.

- ***Find a prepared slide labeled "Fern prothallium young sporophyte w.m."***

Fertilization occurs when sperm cells released from the antheridia swim to an archegonium, and down the **neck canal** to the egg cell. A sporophyte embryo then forms in the archegonium, and eventually grows into a mature sporophyte. Examine the prepared slides that show young sporophytes developing - still attached to a gametophyte.

Horsetails

There is only one genus of horsetails: ***Equisetum.*** We have a living example on display. The genus *Equisetum* is morphologically identical to the fossil genus *Equisetites* from the Carboniferous, some 300 million years ago. If these genera are the same, then *Equisetum* may be the oldest surviving genus of plants on earth -truly a "living fossil".

Horsetail Sporophyte Structure

Horsetails appear at first to be "all stem". But, if you look closely, the stem has 'segments'. Where the segments join, at each "node", you'll find a circle of leaves. These leaves are supplied by more than one vascular strand, and so are called "megaphylls"- despite their small size. The aerial stems branch from underground stems called "rhizomes". And the roots also branch from these rhizomes.

- ***Find a prepared slide labeled "*****Equisetum** ***Mature Strobilus l.s."***

Our plants do not have reproductive structures. These strobili are made up of modified branches, called **sporangiophores,** which bear the sporangia. You should examine the prepared slides of horsetail strobili. These are from a different species of *Equisetum,* so are larger than what you might expect on our living plants.

You may be able to see the **"elators"** on each spore. These structures coil up around the spore under humid conditions, but extend out from the spore as they dry. Elaters help with dispersal of the spores in the wind. We have prepared whole mount slides showing *Equisetum* spores and their elators.

- ***Find a prepared slide labeled "*****Equisetum** ***Spores w.m."*** Some of our slides of *Equisetum* have gone missing -we may adjust by setting up demonstration slides instead.

Figure 15. Spores of *Equisetum*. There are two elaters on each spore. Notice that the ends of the elaters are shaped like paddles. It's common to find elaters on these slides that have separated from their spores.

Lycopodiopsida (Lycopods)

There are three families of Lycopods: **Lycopodiaceae, Selaginaceae** and **Isoetaceae.** We will only be looking at a representative of Selaginaceae, *Selaginella* sp. Molecular data suggest that the lycopods are probably the basal group of the vascular plants.

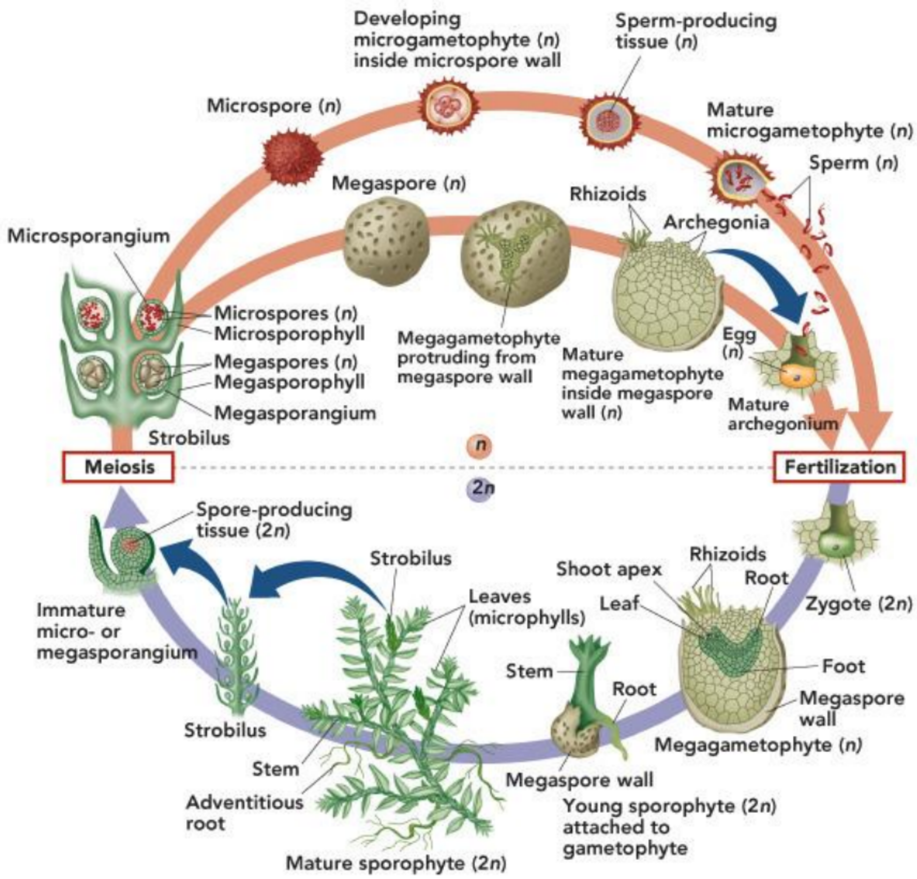

Figure 16. Life-cycle of the Lycopod *Selaginella*.

Selaginella Sporophyte Structure

Selaginella has true stems and leaves, and compared with bryophytes, the plants can be quite large. Lycopod leaves are called **"microphylls"** because they are supplied by only a single strand of vascular tissue (xylem and phloem). The stems contain a central, lobed cylinder of vascular tissue; and there are true, vascularized roots as well.

You should examine the *Selaginella* plants we have on display. You may be able to find **strobili,** located at the ends of some of the branches. There may also be bulbils on some of the plants. Bulbils will also be terminal.

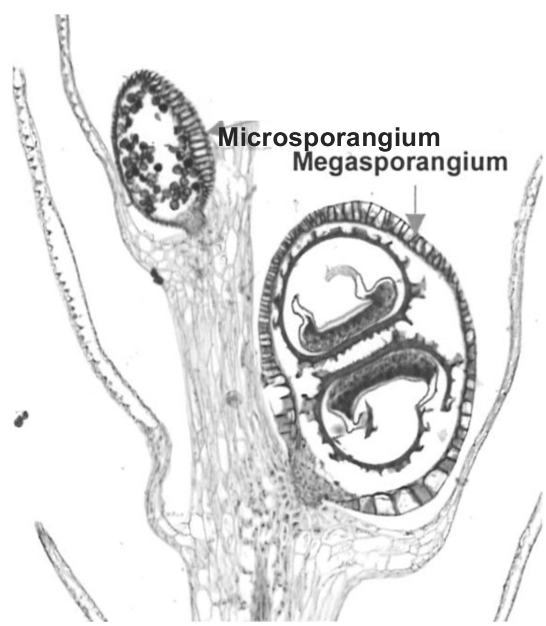

Figure 17. *Selaginella* strobilus and sporangia.

- ***Find a prepared slide labeled "*****Selaginella** ***Strobilus l.s."***

When you study this slide, be sure to distinguish between the Megasporangia and the Megaspores they contain, and the Microsporangia and their Microspores. Megaspores grow into the female gametophyte, and microspores become the male gametophyte.

Lycopod Gametophyte Structure

The gametophytes of lycopods are small and inconspicuous. Some gametophytes develop entirely within the confines of the spore wall, even before the spore is shed *(Selaginella,* isoetes). In *Lycopodium,* the small gametophyte develops on, or just below, the soil surface. In all the lycopod genera, fertilization occurs when sperm cells swim to the egg cell contained in an archegonium.