

UCONN Early College Experience — Biology workshop
 Bryophytes: 400 Million Years of Evolution and into the Classroom
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Note: The plates of black and white illustrations included here are from Crum and Anderson (1984), Mosses of Eastern North America, Volume I. Columbia University Press. The plate below of bryophyte plants and the life cycle are taken from Graham et al. Plant Biology, Pearson Publishing (ISBN-10: 0131469061). All other pictures are by Goffinet and copyrighted.

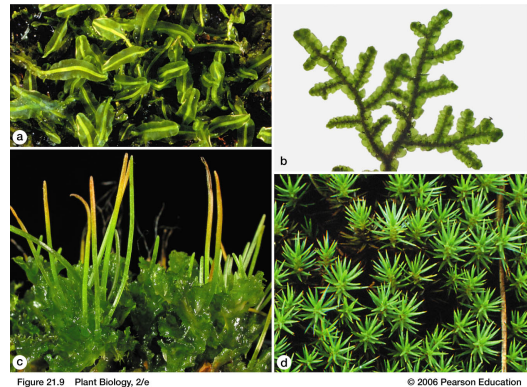
A. BRYOPHYTES DIVERSITY: ON DISPLAY.

Liverworts: *Conocephalum conicum*, *Pallavicinia lyellii*, *Pellia epiphylla* and *Nowellia curvifolia*.
 Mosses: *Thuidium delicatulum*, *Dendroligotrichum dendroides*, *Fontinalis antipyretica*.
 Hornwort: *Phaeoceros carolinianus*.

Some features to distinguish the three lineages of bryophytes.

The vegetative body:

- liverworts: either ribbon shaped, or composed of a stem with leaves (top left and right, respectively)
- hornworts: always ribbon shaped (lower left)
- mosses: always a leafy stem (lower right).



The spore producing body:

- liverworts: urn at the end of delicate translucent stalk, opening by slits rather than a lid (Bottom left)
- mosses: robust stalk with always a leafy stem, with urn opening by a lid (bottom middle)
- hornworts: horn-shaped, and opening along to lines, like a zipper (bottom right).



Liverworts

Mosses

Hornworts

B. FUNDAMENTAL ARCHITECTURE OF MOSSES.

1. Life cycle

The plant: Physcomitrium pyriforme (see plate on next page for details of plant)

About it: This species is widely distributed in North America, occurring on disturbed soil, in gardens, agricultural fields

All land plants have alternations of generations, in fact vertebrates including humans, too, except that in humans just like in seed plants one generation is completely hidden!



Example of alternation generation in ferns: the diploid sporophyte that will grow into the familiar fronds, arises from an embryo held on the maternal plant, here a tiny rosette. Gametophytes of ferns are short lived, so are typically not noticed.

The generations: the gametophyte or gamete producing body and the sporophyte or spore producing body.

The life cycle is an alternation of a gametophytic phase, which is composed of haploid cells, and, following sexual reproduction (the fusion of two gametes), the sporophytic generation, composed of diploid cells. The cycle is completed when meiosis reduces the number of chromosomes per cell to a haploid (original parental) number.

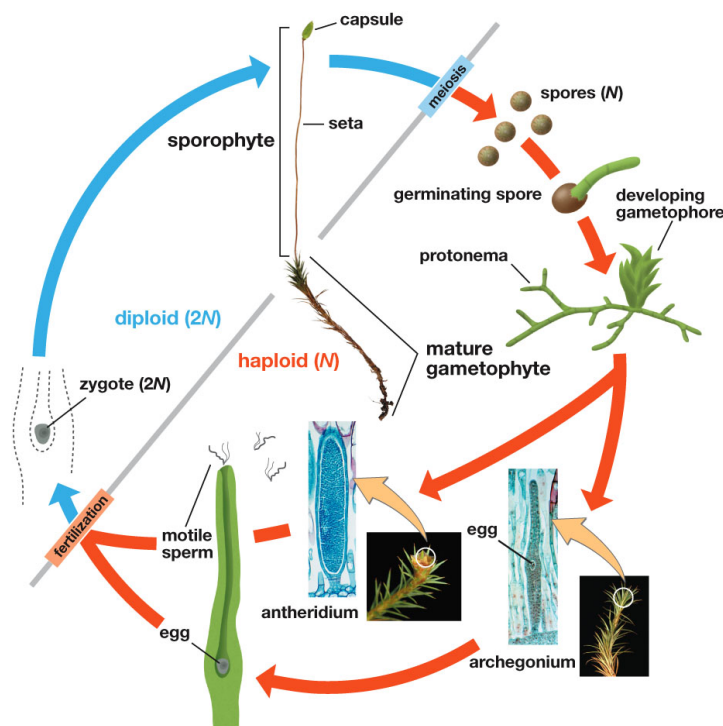


Figure 21.12 Plant Biology, 2/e

Exercise. Consider the sample of *Physcomitrium pyriforme*. This sample was dried and stored, but like all bryophytes it regains its form upon rehydration.

1. Isolate a single leafy plant with an axis bearing an urn attached to it.

These are the two generations: haploid leafy gametophyte and unbranched diploid sporophyte.

The sporophyte develops following sexual reproduction from a fertilized egg, housed in a female sex organ. So the sporophyte is always attached to the plant that produces at least female sex organs. Why at least? Because some mosses produce bisexual plants, with male and female sex organs (Fig. on right: 2 antheridia, male & 4 archegonia, female).



2. Pressing down on the stem with the dissecting needle, use the forceps to pull off the sporophyte. You will notice that it is firmly attached. This reflects the strong contact between the maternal plant and its offspring, which it nourishes through a placenta.

It is worth noting that maternal care, or matrotrophy, is universal among land plants, which are also called embryophytes for they bear an embryo, which acquires nutrients via the placenta.

In some cases other female sex organs, the archegonia (Figure below), with unfertilized eggs can be seen at the base of the sporophyte.



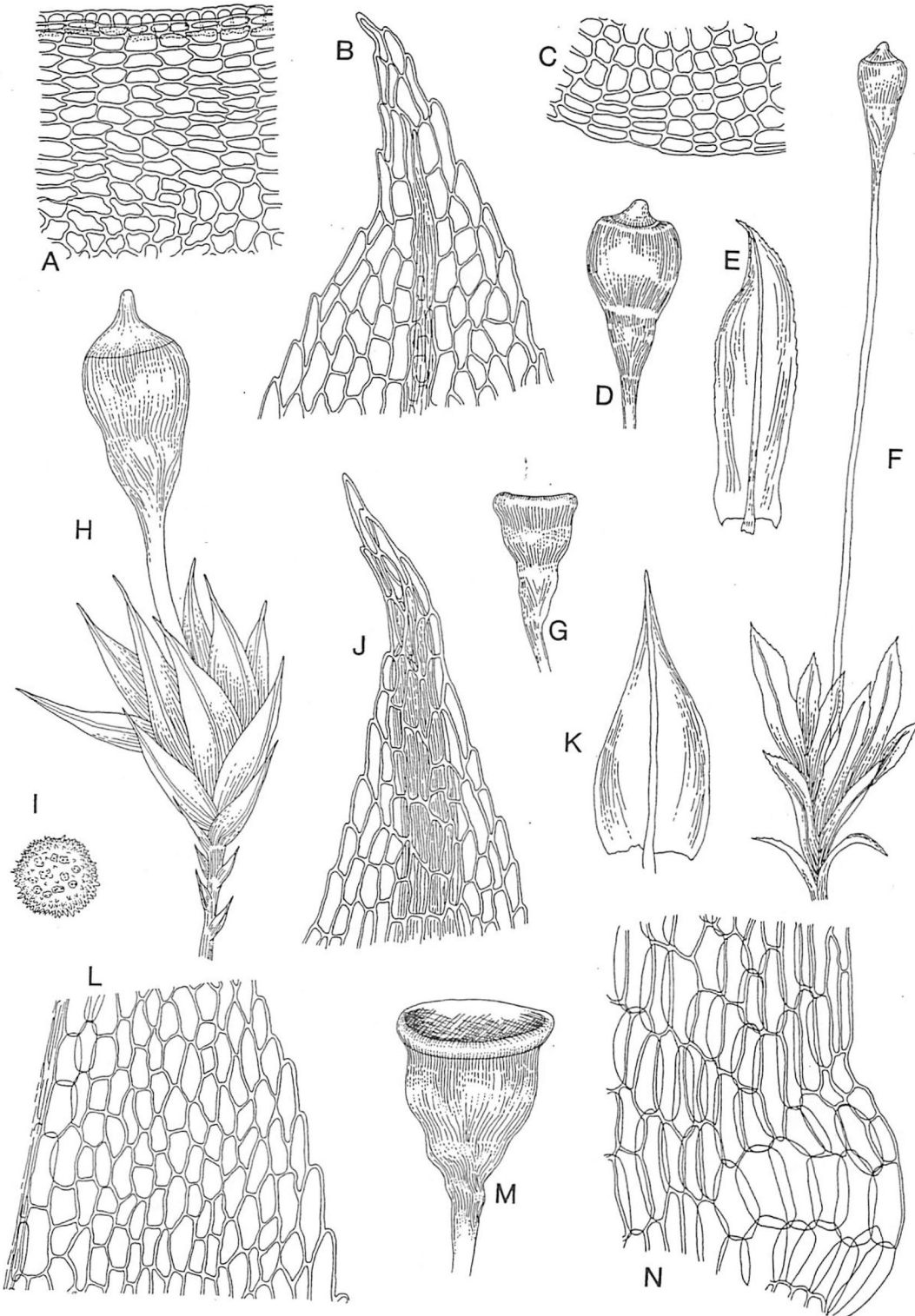


Figure 212 *Physcomitrium pyriforme*. *A*, exothecial cells and portion of annulus ($\times 150$). *B*, cells at leaf tip ($\times 150$). *C*, cells at margin of operculum ($\times 150$). *D*, capsule with operculum, wet ($\times 19$). *E*, leaf outline ($\times 20$). *F*, habit ($\times 7$). *G*, capsule, dry, after operculum is shed ($\times 19$). *H*, habit, "var. *serratum*" ($\times 15$). *I*, spore ($\times 150$). *J*, cells at leaf tip, "var. *serratum*" ($\times 150$). *K*, leaf outline, "var. *serratum*" ($\times 23$). *L*, cells above leaf shoulder ($\times 160$). *M*, capsule, dry and empty ($\times 35$). *N*, cells at leaf base ($\times 133$).

2. The gametophyte

The plant: Plagiomnium cuspidatum

About it: Common moss in New England, in moist lawns; forming light green carpets with spreading runners (like a strawberry plant) and erect branches. The species is often fruiting.

What do you see: a stem and leaves inserted along a spiral line (note that the leaves have no petiole). The stem is erect or creeping with erect branches, and is attached to the substrate by tiny brownish-red filaments, called rhizoids. These rhizoids are genetically homologous (their expression is control by related genes) to the root hairs seen at the tip of roots in seed plants.

Exercise. Take a microscope slide, add a drop of water and place the moss in it. Place this under the dissecting scope.

On the side prepare another slide and add a tiny drop of water.

Under the dissecting scope, remove some of the rhizoids and transfer them to the second slide. Cover them with a slip and observe the rhizoids under the compound scope. Describe them. How many cells wide are the rhizoids? How many cells are stacked?

Clean your slide and cover slip. Place a drop of water on the slide and keep it next to the dissecting scope.

Return to the dissecting scope, to remove some leaves. You can hold the stem down with the dissecting needle and pull a leaf off by pulling it back (downward; see figure). Place the leaf on the slide with the water, cover and transfer to compound scope.



The leaf has a midrib, composed of multiple layers, but the blade itself is unistratose: each cell can be observed, with its numerous chloroplasts.

Scanning the leaf notice the diversity of cell types (see plate on next page)

- a) the margin is strongly toothed
- b) the other marginal cells are linear in several rows, forming either frame or facilitating water movement inside the leaf!
- c) most cells are rather isodiametric with distinct cell walls.

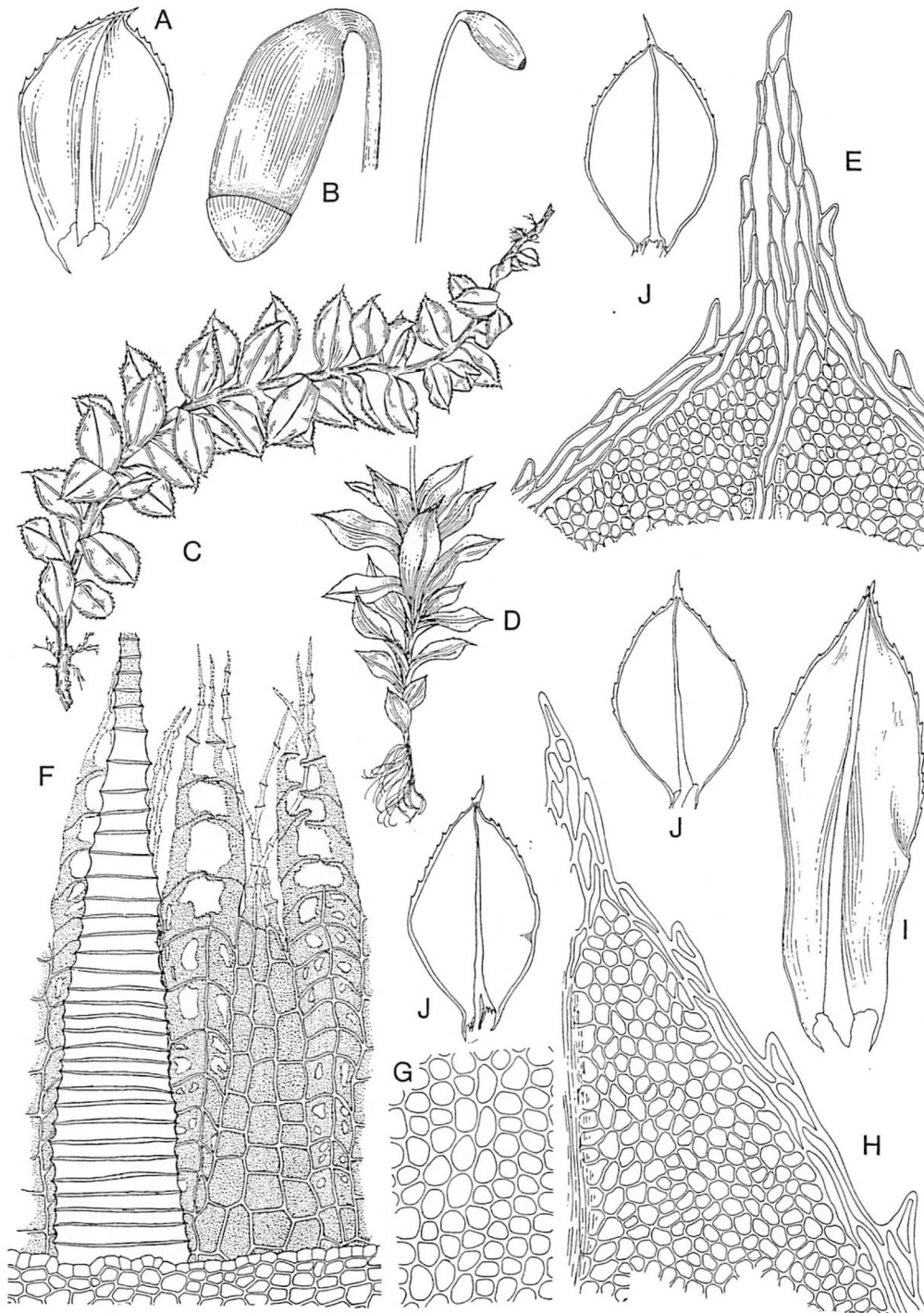


Figure 274 *Mnium cuspidatum*. A, fertile stem leaf outline ($\times 27$). B, capsule, wet ($\times 16$). C, habit, sterile branch ($\times 7$). D, habit, fertile plant ($\times 7$). E, cells at leaf tip, sterile branch ($\times 110$). F, portion of peristome, showing inner face of tooth ($\times 180$). G, median leaf cells ($\times 270$). H, cells at leaf tip, fertile plant ($\times 180$). I, upper leaf outline, fertile plant ($\times 27$). J, sterile leaf outline ($\times 16$).

3. The sporophyte

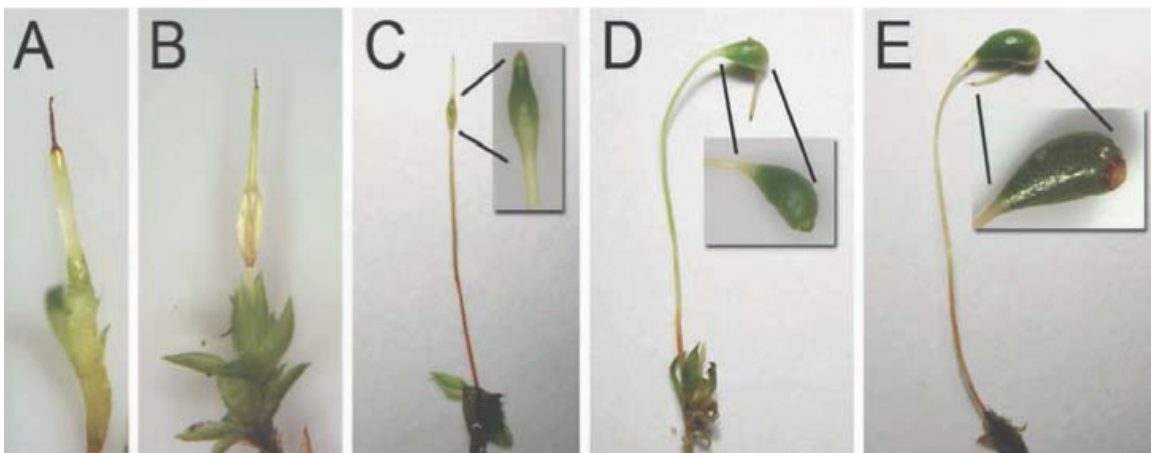
The plant: *Pohlia nutans*

About it: This species has a broad geographic distribution. It grows on soil in moist areas, and is characterized by the short stems, with spear shaped leaves, and the abundant stalks bearing pending capsules.

At this point you will be familiar with the leafy gametophyte, now it is time to take a closer look to the spore producing generation; the sporophyte. As in all the bryophytes, the sporophyte is never branched, and hence always produces only one spore-bearing capsule, and is as always anchored in the maternal plant.

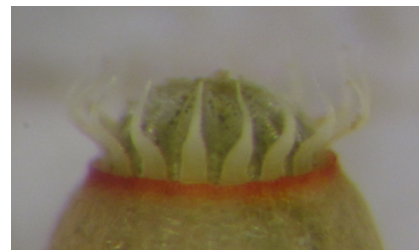
The sporophyte is composed of a long stalk, the seta, with an urn or capsule at its end that will produce the spores, via meiosis. The tip of the capsule is closed by a lid, or operculum. When mature this lid will fall off, to create an opening, the capsule mouth, through which the spores are dispersed.

How does it grow?



Sporophyte development of *Funaria hygrometrica*. Modified from Budke et al. American Journal of Botany 99(1): 14–22. 2012. (A) Sporophyte as a small embryo inside female sex organ. (B) Sporophyte growing and stretching the female sex organ. (C) Sporophyte has ruptured the female sex organs, elongates and starts to enlarge capsule. (D) Capsule enlarging further and spore mother cells being formed. (E) Sporophyte maturing, spores being formed and lid being differentiated above a red ring of cells, the annulus.

Exercise. 1. If your capsule is still closed: Cut the sporophyte off at the base. Take the capsule, and place it in a small drop of water on a slide, hold it between your forceps and with the dissecting needle try to gently pop off the lid, by pushing slightly below the ring at the base of the lid. This should expose a cone of teeth, the peristome (right). This process occurs spontaneously when the capsule matures. Watch what happens when you let this dry out (you can place it on a paper towel, this will speed up the process).



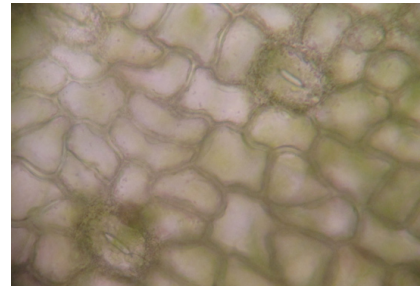
2. Now return the capsule to a drop of water on the slide and cut off the capsule at its base. Hold on to the seta and set aside.

Hold the capsule between the blades of the forceps and cut it with the razor blade lengthwise, in two long halves.

Turn the halves with the inside facing up and holding each half with the forceps push the spore producing tissue out with the dissecting needle. Now turn one half with the outer surface up.

At this point you should have both halves, one exposing the inner side, one exposing the outer side. Cover with a slip. Examine under the compound scope.

Locate the half with the outer surface up. You should see a clear network of cells. These are the epidermal cells of the capsule. They lack chloroplasts. Some of the cells are clearly distinct, the stomatal guard cells.



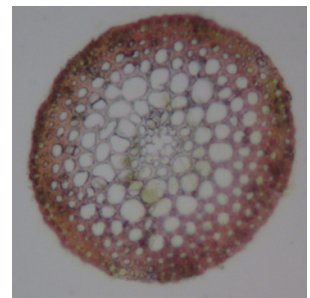
In contrast to liverworts, moss capsules have stomata, but unlike those of seed plants, these may not function for gas exchange but mainly for facilitating water loss, to dry out the spore mass, to allow spores to be dispersed individually!

At the top of the capsule, where the lid came off, are two rings of teeth that lined the capsule mouth. These are the peristome teeth, the structures that regulate spore dispersal (see fig. on previous page).

Examine the other half: depending on the stage of maturation, you can see a mass of spore mother cells, entering or undergoing meiosis, or a sac filled with spores, which will become colored and sculptured with age.

After cleaning your slide, add a small drop of water, and place the seta in it. Holding the seta down with the dissecting needle at right angle, slide the razor blade down the needle to make thin section of the seta (like you would to slide a carrot!). The result is worth the effort!

The seta is made up of distinct tissues include a central axis of conducting cells for water and nutrient transport to the sporogenous tissue! Bryophytes are vascular, too!



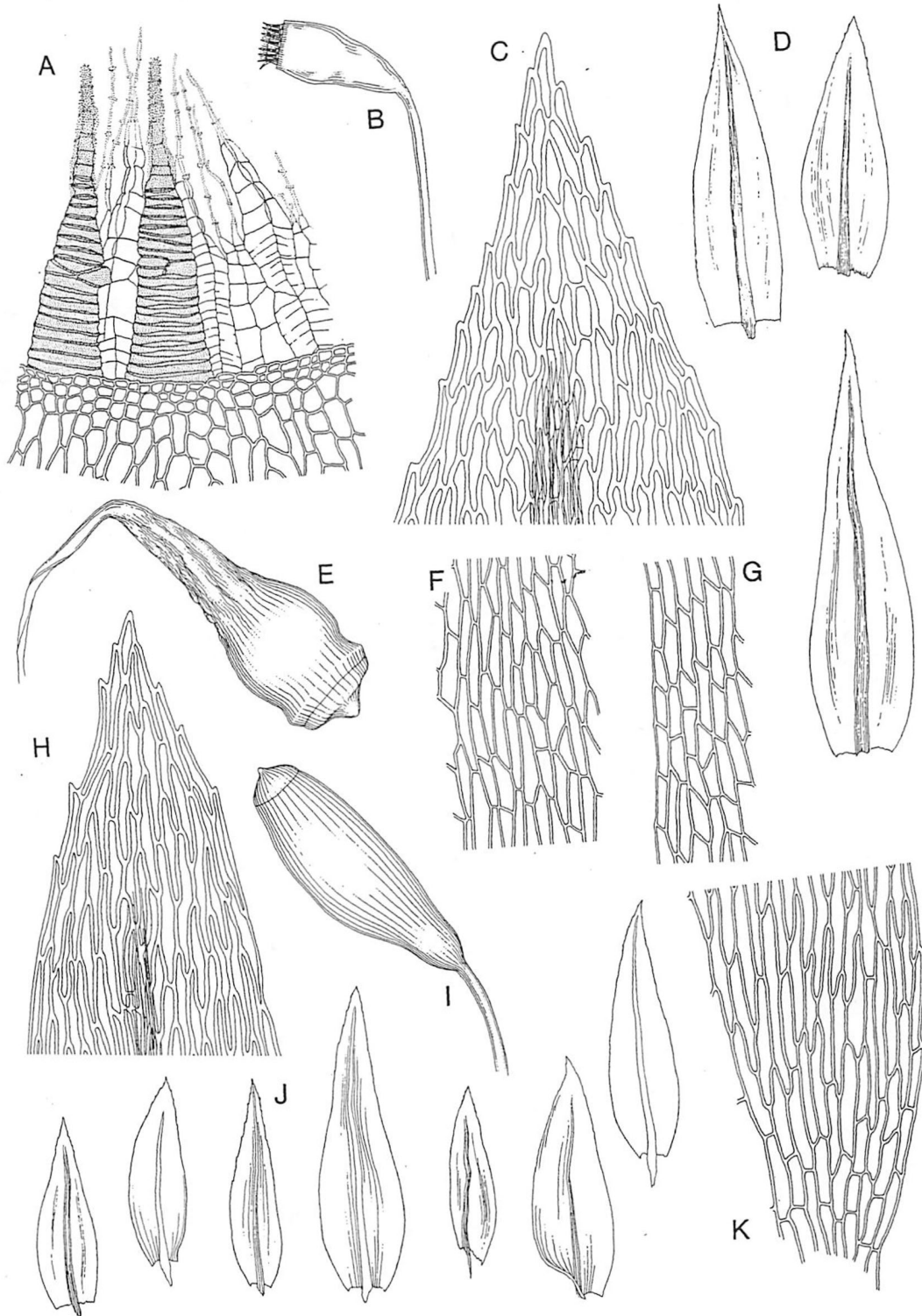


Figure 244 A–G, *Pohlia nutans*. A, portion of peristome, showing 2 outer teeth (focused on inner surface) and 2 inner segments with appendiculate-nodose cilia ($\times 133$). B, capsule ($\times 13$). C, cells at leaf tip ($\times 273$). D, leaf outlines ($\times 25$). E, capsule, dry ($\times 30$). F, median leaf cells ($\times 250$). G, lower median leaf cells ($\times 250$). H–K, *Pohlia cruda*. H, cells at leaf tip ($\times 273$). I, capsule, wet ($\times 11$). J, leaf outlines ($\times 25$). K, cells at leaf base ($\times 270$).

Bryophytes in the classroom

Today's exercise illustrated that mosses can be used in the classroom to easily illustrate

- the concept of alternation of generations, which characterizes all land plants and in fact animals, too!
- maternal care in plants; matrotrophy is shared by all land plants: they all hold on to their embryo to nourish it and give it a head-start!
- plant cells (cells with a rigid cell wall and chloroplasts – use leaves that are unistratose)
- stomata: a major adaptation in land plants to ultimately overcome the problem of controlling water loss while absorbing CO₂
- movement in plants: the peristome of mosses, is composed solely of cell wall material; movement in plants involved turgor and modified cell walls, or differentially thickened walls (cells will shrink when losing water, along an axis determined by the weaker cell walls!).
- the concept of descent with modification, a central concept in evolution, by focusing on the evolution of generations: the gametophyte of mosses vanishes inside the flower and the unbranched sporophyte of mosses becomes a tree!
- role the cuticle: vegetative moss bodies typically lack a cuticle, hence absorb and lose water through their entire surface; the independence of water in the soil, allows them to grow on trees, and rocks! Few seed plants can do that!
- desiccation tolerance: unlike most seed plants that avoid desiccation by enter a dormant state during dry periods, many mosses can tolerate complete desiccation, stay dry and regain physiological activities within minutes or hours of rehydration.

These are just a few examples of our bryophytes could enter the classroom and be used to discuss or illustrate major concepts in biology.