A. Introduction

The shoot consists of stems and leaves. It is quite modular in its construction. A shoot is made up of repeated units known as PHYTOMERES (see Figure 1 below). Inspect the figure and note the basic rules of plant architecture.

Fig. 1.

Leaves are always attached to what? What is the point of attachment called?

Where are AXILLARY BUDS positioned?

What develop from AXILLARY BUDS?

From what do stems and leaves ultimately develop?

Why are more distal internodes shorter than those less distal?

Leaves function in what?

Stems function in what?
The arrangement of leaves on a stem is referred to as **PHYLLOTAXY** (*phyllo* is derived from Greek for “leaf”, and *taxy* is derived from Greek/Latin for “arrangement” or “order”).

ALTERNATE phyllotaxy is where leaves are attached one per node. Some distinguish between subforms of alternate, such as *distichous* (where the leaves are arranged in one plane) and *spiral* (where leaves are spirally arranged along the stem) (Fig. 2).

OPPOSITE phyllotaxy is where there are two leaves per node (Fig. 2).

WHORLED phyllotaxy is where there are three or more leaves per node (Fig. 2).

**Revisit Fig. 1. What is the phyllotaxy of the diagram in Figure 1?**

**Special notes:**
Sometimes the leaves are clustered so close together on a stem that the plant is said to have its leaves arranged in a **ROSETTE**. In this case, there is an underlying phyllotaxy such as alternate, opposite or whorled, but it may be difficult to discern.

Sometimes these rosettes can appear at the ends of the stems (i.e., terminal rosettes). Other times, the rosette of leaves is borne on an extremely short stem at the ground level, and then the leaves are said to be **BASAL** since the rosette of leaves appears to be positioned at the “base” of the plant.

![Leaf Arrangement](image.png)

**Fig. 2. Three categories of phyllotaxy. Illustrations by MU student Jess Landis (’08).**

Each student group of 4 has a plant of *Setcreasea* (purple heart) and *Rhoeo* (oyster-plant) on their bench. They are very closely related (both are in the Spiderwort & Wandering-Jew family) yet look rather different in the arrangement of leaves.

*Does Setcreasea have nodes? Internodes?*
*Does Rhoeo have nodes? Internodes?*
*Rhoeo is said to have a ROSETTE habit. What does this mean?*
*What is responsible for the difference in the apparent arrangement of the leaves between Setcreasea and Rhoeo?*
C. Dissection of shoot apex (living material)

Find the shoot apex and, in particular, the dome above the shoot apical meristem of sunflower (Helianthus) seedlings. This is VERY small. To do this, pair-up with another student and a seedling on your bench, and dissect away the larger leaves (cut them gently at their petioles) surrounding the shoot apex.

What do you see with the naked-eye? With the dissecting scope? With the compound scope (with razor thin longitudinal median section mounted on a slide)? *Draw what you see.*
D. Investigations of Tobacco Shoot Structure (living material)—dissecting scope

Work in pairs for this exercise.

D1. With the TOBACCO plant (a typical dicot), answer the following questions.

How are the leaves oriented on the stem (i.e. opposite or alternate)

What relationship do you see between the leaf size and its position?

Locate the shoot apex.

What structures surround the apex?

How does internode length change from the base to the tip?

D2. Use a razor blade to cut a portion of the stem in half along the longitudinal axis (from base to tip). This segment should cut through at least ONE NODE so that you actually cut the leaf petiole in half also (see Fig. 3). A segment about one inch long will be enough. Coat the cut edge with Toluidine Blue, let sit for 30 sec, then rinse off the stain with water. Use the dissecting scope to make the following observations of the longitudinal section.

Make a sketch of the longitudinal section. Include the points of attachment for the petioles of the leaves. Label the structures mentioned below:

Locate the presence of ground, dermal and vascular tissues.

Locate the vascular bundles and follow them as best as possible

How do you know these are vascular tissues?

Compare the vascular cells to the ground and dermal cells.
Can you see vascular linkages from the stem to the leaves?

D3. Make a variety of cross sections of the tobacco stem. Make at least one cross section that cuts through a node and petiole and at least one cross section that is in the middle of an internode. With the dissecting scope, compare what you see in these two sections after staining them with the Toluidine blue.

Draw the cross sections and label the vascular tissue in each.

Which section has a continuous ring of vascular tissue?

Label any gaps in the vascular tissue.

What kind of ground tissue is the central “pith” of the stem made of?

E. Investigations of Rhoeo Shoot Structure (living material) – dissecting scope

Work in pairs for this exercise.

E1. Now using Setcreasea (a monocot), identify and label your drawing with all of the structures described including the leaf, stem, node, internode, axil, axillary bud, and phytomere.

How are the leaves oriented on the stem (i.e. opposite or alternate)?

What relationship do you see between the leaf size and its position?

Locate the shoot apex.

How does it compare to the tobacco?

How does internode length change from the base to the tip?

E2. Prepare a thin cross section of a Setcreasea INTERNODE and stain it with the Toluidine blue. Locate the vascular tissues and label them in your drawing.

How does the organization compare to tobacco?

Are there any vascular bundles in the pith?

Make sure you understand the organization before moving on!
E3. Use a razor blade to cut a portion of the stem in half along the median longitudinal axis (from base to tip). This segment should cut through at least ONE NODE so that you actually cut the leaf petiole in half also (see Fig. 4). A segment about one inch long will be enough. Coat the cut edge with Toluidine Blue, let sit for 30 sec, then rinse off the stain with water. Use the dissecting scope to make the following observations of the longitudinal section.

Make a sketch of the longitudinal section.

Locate the presence of ground, dermal and vascular tissues.

Locate the vascular bundles and follow them as best as possible.

Compare the longitudinal section to the cross section of the stem. Use the cross section to orient yourself when observing the longitudinal section.

How does the organization of the vascular tissues compare to tobacco?

Can you see vascular linkages from the stem to the leaves?
F. Comparative Anatomy of Stems

The organization of the stem has a lot of similarities with the root. The dermal, ground and vascular tissue systems are readily recognizable and are made of the same types of cells. In the following exercises, you will become familiar with the differences between dicot and monocot stem organization as well as be able to relate this new information back to the root anatomy for comparison.

**F1. Typical Dicot: Helianthus stem cross-section (cross-section abbreviated as “c.s.”)**

- Things to see & do:  
  - dicot organization of 3 tissue systems (as per Fig. 5).
  - types tissues (i.e., parenchyma and collenchyma) in the ground tissue.
  - diversity of cell types in the xylem and phloem. Note the fibers capping the phloem-sides of the vascular bundles.

*Helianthus* (sunflower) exhibits the typical dicot arrangement of the 3 tissue systems (i.e., dermal, ground, & vascular tissue systems). Observe the prepared slides and locate each of the labeled features in the Fig. 5.

Notice that each vascular strand is separated from its neighbor by ground tissue (where pith and cortex come into contact).

Now focus on a vascular bundle. Locate the xylem (water, mineral) and phloem (sugar solution) conducting tissues in each bundle. Note their appearance (e.g., cell types and forms). Note that xylem is a complex tissue.

Cross-sections do not show you the wall thickening patterns of xylem cells. But they do show you how xylem cells differ in girth (diameter). Metaxylem cells have larger diameters than protoxylem cells and they are further away from the pith than are protoxylem cells.
Draw your dicot cross section and label the different vascular tissue cell types. Include labels for the ground tissue (parenchyma, collenchyma, sclerenchyma) the cortex, pith and epidermis.

How does the stem organization compare to the root organization of a dicot?

How does dicot stem organization compare to monocot root organization?

What is the layer of cells surrounding the vascular bundle?

*Helianthus* demonstrates the VASCULAR CAMBIUM. Locate and label it. This region is between the xylem and phloem cells.

Locate and label the fibers capping the phloem region.

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Fig. 6. *Helianthus* (sunflower) vascular bundle, close-up. The blue stain is picking up the cellulose of the primary cell wall. The red stain is picking up the lignin of the tracheary elements and fibers.
F2. Typical Monocot: *Zea (maize)* stem cross-section (cross-section abbreviated as “c.s.”)

Things to see & do:  
- monocot organization of 3 tissue systems.
- types of tissues (i.e., parenchyma and collenchyma) making up the ground tissue.
- all structures labeled in Fig. 7.

*Zea* (corn, maize) exhibits the typical MONOCOT arrangement of the 3 tissue systems. Observe the prepared slides.

*How is this organization different from that of the typical dicot?*

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**Fig. 7.** *Zea* (maize) vascular bundle, close-up. The blue stain is picking up the cellulose of the primary cell wall. The red stain is picking up the lignified secondary cell walls of the xylem vessels and fibers.
F3. Zea (maize) stem longitudinal-section (long-section abbreviated as “l.s.”)

Things to see & do:  -organization of 3 tissue systems.
-types of tissues (i.e., parenchyma and collenchyma) making up the ground tissue.

Can you discern the 3 tissue systems in the long section?

Relate organization perceived in long-section with that seen in cross-section.

Now, look for the xylem and phloem in the vascular bundles.
Xylem is closer to the center of the stem than phloem.
G. Basic Leaf Morphology

Basic leaf structure includes the **PETIOLE** that attaches the leaf to the stem (leaves are referred to as “sessile” when there is no petiole), the **BLADE** which is the flattened portion of the leaf attached to the petiole, the **MIDRIB** or midvein which is the thickened longitudinal rib of many leaves, and will serves as the vascular link to the stem, smaller **VEINS**, and the **MARGIN** or outer edge of the leaf. Be aware of these terms when examining the plants in today’s lab.

Leaves will vary from species to species in their **VENATION** (vein pattern), **MARGIN** (outer edge), **COMPLEXITY** (shape and dissection of the blade), and **ARRANGEMENT** (positioning on the stem). Refer to the **Photo Atlas Fig 9.61** to review what these terms mean.

Fig. 8, left. Basic leaf structure.

Fig. 9, below. Some terms used to describe the complexity of leaves. Illustrations by MU student Jess Landis (’08).
Fig. 10. There are three primary ways that the primary veins of leaves can be arranged. Parallel venation is typical of monocotyledonous plants, pinnate or palmate typical of dicotyledonous plants. Illustrations by MU student Jess Landis ('08).

Fig. 11. Leaf margins come in various forms. Can you think of trees you know that match each category shown here? Illustrations by MU student Jess Landis ('08).
Fig. 12. Leaves can be attached to the stem in various ways. The differences shown above really reflect differences in the form of the leaves, and have nothing to do with phyllotaxy (aka leaf arrangement). Illustrations by MU student Jess Landis ('08).

G1. Work in groups of 3-4. Move around the room and examine the leaves of the 12 PLANT SPECIMENS provided and determine the venation, margin, complexity, and arrangement of each. In addition, be sure to notice the parts of the leaf and make how the shoot is organized (i.e. the phytomere). Record your observations of the leaf morphology below:

**Leaf Morphology Observations:**

<table>
<thead>
<tr>
<th>Plant</th>
<th>Venation</th>
<th>Margin</th>
<th>Complexity</th>
<th>Arrangement (phyllotaxy)</th>
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<th>Plant</th>
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H. Basic Leaf Anatomy

H1. **PREPARED SLIDE:** Typical dicot (lilac) leaf cross-section (c.s.)
Observe the anatomy of a typical DICOT leaf using the typical dicot leaf cross-section (c.s.).
Please note that figure 13 is a schematic illustration of an idealized leaf. Use figure 13 to orient yourself on the leaf cross-section slide.

![Leaf Cross-Section Diagram](image)

In the space provided, DRAW your observations.
Label all of the tissues, cells indicated in Fig. 13.
Identify and label the Xylem and Phloem in the vascular bundle.
Indicate the presence of any AIR SPACES.
Why would air spaces be important in a leaf?

I. Basic Leaf Anatomy

I1. **Living Tissue:** Place a drop of water on a clean slide. Make a 0.5 cm² upper epidermal peel from a leaf of *Rhoeo* (spiderwort family). Place the peel face up on the drop, then place another drop over it and cover with a cover-slip. Repeat with the lower epidermis. Observe the tissue using the compound light microscope at 20X or higher for good comparison.

Do you see stomata with the compound scope?

Draw a region of the **UPPER** leaf epidermis that contains both guard cells and normal epidermal cells.

Draw a comparable region of the **LOWER** leaf epidermis.

**Note the frequency of the guard cells on the upper surface and compare it to the lower.**

Are stomata more frequent on one surface than the other?
I2. **PREPARED SLIDE:** Return to the Typical dicot leaf in x-section.

Look for stomata in x-section.

*Draw and label the stomata plus some of the adjacent epidermal cells.*

![Stomata and Epidermal Cells](image)

*Fig. 14. Cross section example of guard cells surrounding a stomatal pore with subsidiary cells on each side*

Locate the ground tissue (that tissue other than the epidermis and vascular bundles, in the middle of the leaf). This ground tissue in the leaf is called “MESOPHYLL” and it may be differentiated into “SPONGY MESOPHYLL” and “PALISADE MESOPHYLL” in most horizontal leaves (i.e., those with upper and lower surfaces). The palisade mesophyll consists of columnar and more densely spaced cells packed with chloroplasts. The spongy mesophyll also has chloroplasts and its most conspicuously not as densely packed as the palisade mesophyll (i.e., there are more intercellular air spaces in the spongy mesophyll).

I3. **Rhoeo, free hand cross-sections.**

Using two pieces of styrofoam cups to gently but securely “sandwhich” a section of leaf (ask instructor for technique details), use sharp razor blade to make razor-thin cross-(transverse)-sections of the leaf. Make several and float them into a small petri-dish of water to keep them moist until you are ready to mount them. Then take the thinnest section(s) and mount them onto a slide.

*Is the mesophyll differentiated into palisade and spongy mesophylls in this species? If so, relate this to the upper vs. lower sides of the leaf. Explain any pattern in terms of functional anatomy and ecology.*
J. Leaf Adaptations: Mesomorphic Leaves (typical leaf anatomy)
Review your observations of the typical dicot leaf. Make a note of how the PALISADE PARENCHYMA and the SPONGY PARENCHYMA are arranged.

How many layers do you typically see for this type of leaf?
Also note the appearance of the epidermis and the ground tissues.
Use your observations to compare this type of leaf to the leaves in Sections E – I.

K. Leaf Adaptations: Xeromorphic (arid leaf anatomy)
K1. Observe and DRAW the xeromorphic leaf examples: *Nerium oleander & Ammophila* (“beach grass”) in cross-section. These types of leaves are specially adapted to growth in harsh, arid environments. *Nerium* has fine examples of trichomes and *Ammophila* demonstrates stomata.

How do these leaves differ from Lilac?
Label your drawing with the typical tissues, cells.
Label the STOMATAL CRYPTS and CUTICLE.
What purpose do these structures have?
Identify and label the trichomes and stomata.
Where are they found?

How is the cuticle different from Lilac?

How does the Epidermis differ?

Is there any change in the basic organization of the ground or vascular tissues?

L. Leaf Adaptations: Hydromorphic Leaves (water leaf anatomy)
L1. Observe and DRAW the hydromorphic leaf example: *Nymphaea odorata*, “water-lily” in cross-section. This type of leaf is adapted for aquatic habitats and often is submerged in water.

Draw the cross-section of the water lily.

Label all of the typical tissues and cells.

Label the intercellular spaces.

How does this type leaf differ from the previous two?

How would the organization of the ground tissue help this type of plant survive?
Do you see any SCLERIDS in the mesophyll?  
What might their purpose be?  

How does the mesophyll tissue differ from xerophytes?  
Are there any differences in the organization of the 
Ground and vascular tissues compared to mesophytes?

M. Unknown Leaf Identification Study: Determine if Meso-, Xero-, or Hydrophyte

M1. Use the prepared slide labeled “Leaf-types cross-section” for this part. This slide has examples of all three types of leaves looked in the previous sections. If professor asks, on a blank page DRAW and LABEL each cross-section as the type of leaf it represents (there is space on back page). Label the typical structures including: epidermis, ground, vascular tissues, mesophyll, palisade parenchyma, spongy parenchyma, bundle sheath cells, xylem and phloem. In addition, LABEL the structures and differences that are associated with that type of leaf.

N. Variations of leaves: MONOCOTS with Grass-like (vertical) leaves
The leaves of monocots, like grasses, have a slightly different organization but still contain all of the primary tissues one would expect. Consider the differences in comparison to the Lilac leaf.

O1. Observe the cross-section of a typical monocot (in this case grass) leaf. Draw and label the cross-section with the typical leaf tissues and cells. Include the specialized features.

How is the ground tissue organized differently from the dicot leaf?  
What is missing?  
Suggest a reason for the missing layer.

Distinctive to many grasses are BULLIFORM CELLS? Find these and suggested an adaptive function.
O. Morphological Plasticity: Sun vs. Shade Leaves
Water availability is not the only environmental factor that influences the development of the leaf. Changes in the basic organization can also be seen based on the INTENSITY of light the leaves develop in. In other words, leaves that develop in full sunlight and leaves that develop in shade have differences in their anatomy. Interestingly, these types of leaves can be found on the same plant.

N1. Observe the prepared slide: *Sambucus* (elderberry) sun vs. shade leaf cross-section. Compare the sun and the shade leaves under the microscope, and fill in the table below.

<table>
<thead>
<tr>
<th></th>
<th>Sun</th>
<th>Shade</th>
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</thead>
<tbody>
<tr>
<td>Thickness of cuticle (thin, thick)</td>
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<td></td>
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<tr>
<td>Amount of palisade mesophyll; (organized in single or multiple layers)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chloroplasts per unit area (density is High or Low)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leaf thickness (thin or thick)</td>
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</table>

Consider what this tells you about the function of those leaves.

Why might the differences be beneficial for leaves growing in the shade or in full sun?

What part of a plant would you expect to find either of these leaves in?

P. Leaf Abscission: (Optional)
Many woody species of plants lose their leaves in the fall through a process known as ABSCISSION. This process results in the degradation of a SEPARATION LAYER and the suberization of a PROTECTIVE LAYER.

P1. Observe the prepared slide of *Salix* leaf abscission.

Identify the separation and protective layer.

Which layer will form the LEAF SCAR recognizable on the stem after abscission?

Why are both layers important in the abscission process?

Are any cells suberinized or lignified?
Optional Assignment (as per instructor’s fancy): Comparative Analysis of Leaf Types
Use the prepared slide labeled “Leaf-types cross-section” for this part. This slide has examples of all three types of leaves looked in the previous sections. On the last page of this handout, use the space provided to DRAW and LABEL each cross-section as the type of leaf it represents. Label the typical structures including: epidermis, ground, vascular tissues, mesophyll, palisade parenchyma, spongy parenchyma, bundle sheath cells, xylem and phloem. In addition, LABEL the structures and differences that are associated with that type of leaf.

**MESOPHYTE:**

**XEROPHYTE:**

**HYDROPHYTE:**