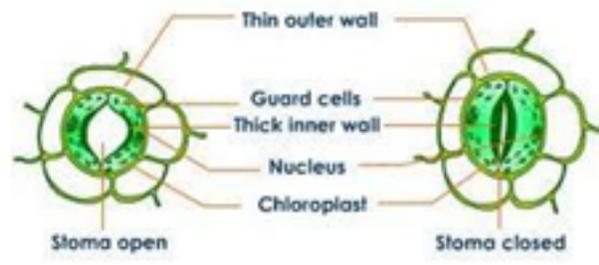


Developed by:
Surinder Kaur, Science Leadership Academy

Plant Stomata Lab



Introduction and Background:

The epidermis of a leaf has microscopic openings, which are the “pores” of the plant’s skin on the undersides of leaves called stomata (*sing.* stoma or stomate). Stomata allow the exchange of gases such as oxygen and carbon dioxide between the plant and the outside environment. Water vapor is also released through the stomata in a process called transpiration. The rate of gas and water vapor diffusion is regulated by two guard cells. The shape of the guard cells will change when water enters the plant by the process of osmosis. When the stomata are open, diffusion rates of carbon dioxide, water vapor, and oxygen are greater. Therefore, when water exits, the guard cells relax, the stomata close, and diffusion rates are reduced. The closing of the stomata help the plant by preventing dehydration due to loss of water vapor, and helps with maintaining the water necessary for photosynthesis and the homeostasis, or internal balance of the plant. It is also important to mention that this process is a crucial step in the Earth’s water cycle. Generally, stomata are open during the day and close at night. Functioning of the stomata could be affected by light intensity, stress (disease, insects, etc.), climate, and/or latitude.

The focus of this lab is to learn how to prepare a cast of leaf stomata and to compare stomata distribution on the top (upper surface) and bottom (lower surface) of a leaf.

Materials:

Compound Microscope

Microscope slides

Clear tape (shiny, not translucent)

Clear fingernail polish (not strengthener)

Centimeter ruler (straight edge for labeling)

Various leaf specimens- may be live, or dead (dried flat); pin oak is an excellent species for observation

Guiding Questions:

1. Where would you expect to find more stomata, on the top or underside of a leaf?
2. Will the number of stomata vary among different plant species? Why might this be the case?
3. Would you expect stomata to be the same size and shape in different plant species?
4. What environmental conditions might cause guard cells to open or close the stomata?

Vocabulary:

1. **Diffusion** is the movement of particles from an area of high concentration to an area of low concentration; a natural result of kinetic molecular energy.
2. **The epidermis** is a single-layered group of cells that covers the leaves, flowers, roots and stems of plants.
3. **Guard cells** are the paired cells in the epidermis of a plant that control the opening and closing of a stoma of a leaf.
4. **Homeostasis** is the regulatory process in which an organism regulates its internal environment.
5. **Osmosis** is the movement of water or another solvent through permeable membranes from an area of higher water concentration to an area of lower water concentration.
6. **A stoma or stomate** is a minute pore in the epidermis of plant leaves or stem. (*plural stomata*).
7. **Transpiration** is the process by which water that is absorbed by plants, usually through the roots, is evaporated into the atmosphere from the plant surface, through the stomata.

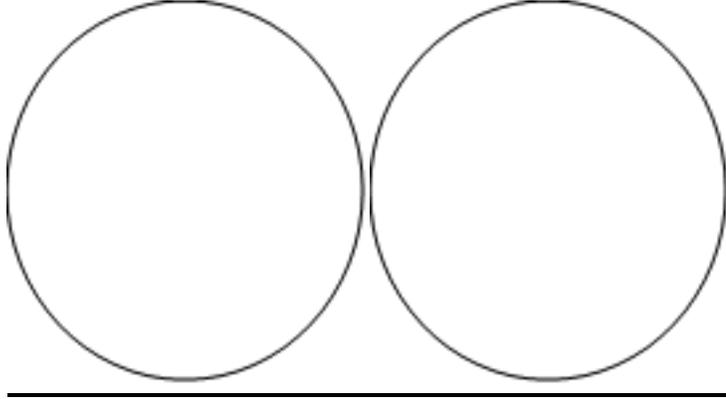
Procedure:

1. Select a leaf. Be sure it is dry. To prepare cast of lower (abaxial) leaf surface, paint a small rectangular area of the underside of a leaf with nail polish (approximately 1 cm x 2cm). Allow the area to dry completely, usually at least 5 minutes. No longer than approximately 10-15 minutes should be allowed because the polish might be difficult to remove without damaging the leaf if the drying time is extended.
2. Repeat the same procedure from in step 1 with the upper (adaxial) surface of the same leaf.

3. While waiting for the polish to dry, label the end of two clean microscope slides with a small (1-2 cm) piece of colored or masking tape with your initials, date, u. (upper side) or l. (lower side).
4. Cut a piece of clear tape approximately 4.0 cm in length. Fold a 1-2 cm portion of the tape over onto itself to use as a "handle." Once the polish is dry, place the sticky surface on the nail polish cast and press gently with your finger and rub back and forth about 5 seconds. Using the "handle," carefully pull the nail polish cast from the leaf surface, taking care not to tear the leaf. You should observe some of the leaf epidermis (skin) on the tape.
5. Place the cast (sticky side down) in the central area of the labeled slide. Press gently in place. The adhesive on the tape should mount the cast onto the slide. **DO NOT REMOVE THE TAPE OR ADD A COVER SLIP.**
6. Starting with the lowest objective (4x, bring the leaf epidermis into view and look for stomata. Next change to the 10x objective where stomata if present should be very visible. The stomata should look like round or oval circles with slits in the center (see photos at beginning of lab sheet). Try to observe if stomata appear open or closed. Make a sketch of at least a few stomata and surrounding plant tissue at this magnification and try to count how many stomata you can observe in your field of view for the 10x objective. Write the total magnification of the observed cells in the space provided on the Data sheet. **Remember to multiply the power of the objective by that of the eyepiece (10x) to obtain total magnification.** Finally, try to use the 40x objective to obtain a detailed view of the stomata. This power might be more difficult to focus and observe clearly. Ask your teacher for help.
7. Observe the prepared slide of the upper side of the leaf following the procedures in 6.
8. Remember you might not see any stomata on the upper side of the leaf, but will observe epidermal cells.

Student Lab Data Sheet

A. Sketch (**upload a picture**) a small section of both the under and upper sides of the leaf you have observed using the 10x objective in the circles below. If applicable from results, label several of the **stomata** with **guard cells** that you might observe and **epidermis** in either view. Use a ruler to draw lines to structures.



Underside of Leaf
Magnification _____ x

Upper Side of Leaf
Magnification _____ x

- B. Approximately many stomata did you observe in your field of view at 100x on the under side of the leaf? _____
- C. Approximately many stomata did you observe in your field of view at 100x on the upper side of the leaf? _____
- D. Now obtain class data. What is the average number of stomata observed on the underside of the leaves observed by the class? _____
- E. What is the average number of stomata observed on the upper side of the leaves observed by the class? _____

Analysis:

1. How do the guard cells assist the stomata? How do guard cells open and close stomata?
2. Which side of the leaf had more stomates?
3. Explain why leaves might have more stomata on one side. (You may research your answer for more information).
4. At what time of day would stomata be closed and why?
5. Why does the lower epidermis have more stomata than the upper epidermis of a leaf?
6. Define transpiration.
7. What substance enters the leaf and what substance exits the leaf through the stomata?
8. What part of your body does the stomata of the leaf most closely resemble. Why do you think this?
9. How are stomata related to the process of photosynthesis? Explain using the photosynthesis equation in your answer.
10. What was the most difficult part of this lab and why?

Acknowledgements

The contents of these digital library curricula were developed by teacher participants in the Research Experience for Teachers program at the Center for Engineering MechanoBiology under National Science Foundation STC grant no. (CMMI): 15-48571. However, these contents do not necessarily represent the policies of the National Science Foundation, and you should not assume endorsement by the federal government.