

## **Stomata Investigations**

Peggy O'Neill Skinner  
The Bush School  
Seattle, Washington

### **Overview**

In this lab you will learn a very simple technique to make a cast of the outer surface of plant tissues. Using your cast and a microscope, you will see different types of epidermal cells. After identifying the structures that define a pair of guard cells and their accompanying stoma, you will design an experiment to test the distribution and/or function of stomata in land plants.

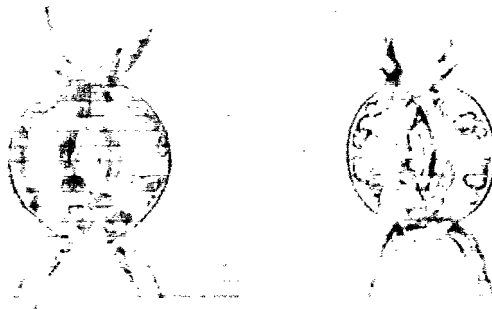
### **Background**

Stomata are gaps in the epidermis of plant tissues that are bordered by a pair of guard cells. Stomata are the avenue of gas exchange for the plant. They are also the avenues of water loss through the process of transpiration. The mechanism that controls the opening or closing of stomata is based on water potential of the guard cells compared to their surrounding cells or environment. Stomata open as a result of water moving into the guard cells. That process is regulated by the active transport of  $K^+$  ions into the guard cells. The active transport of  $K^+$  ions decreases the water potential of the guard cells, causing them to take up water. The increased water in the cells causes the guard cells to change shape and opens the stomata. Stomata usually are open during the day and closed at night, balancing the need for photosynthesis and water conservation.

### **Procedure**

1. Obtain a leaf and dry it if necessary. Paint a small section (not more than  $1\text{ cm}^2$ ) of the underside of the leaf with clear fingernail polish. Let the polish dry completely (510 minutes).
2. Place a small piece of clear adhesive tape onto the painted portion of the leaf. Press gently. Lift the tape off the leaf. The patch of fingernail polish should adhere to the tape.
3. Place the tape and fingernail-polish cast sticky side down onto a clean microscope slide. There is no need for a cover slip because the tape keeps the sample aligned.
4. View the cast under a microscope. High power (400x) is the best magnification for viewing detail, but low power (100x) may be the best magnification for counting the number of stomata in a field of view.

5. Observe the stomata that appear as the space between pairs of guard cells. The stomata may be open or closed. See diagram.



Courtesy of Graham Kent

6. Count the number of stomata in one field of view under low power (100x). If there are too many to easily count under low power, switch to high power (400x). Record your data and move to two additional fields of view to count the stomata. Average your three counts.

Microscope magnification: \_\_\_\_\_

Number of stomata: Trial 1 \_\_\_\_\_ Trial 2 \_\_\_\_\_ Trial 3 \_\_\_\_\_ Average \_\_\_\_\_

7. Calculate the average density of stomata per mm<sup>2</sup> using the following technique:

**Low power (100x)** field of view diameter = 1.760 mm (verify this with a millimeter ruler and your microscope).

$$\text{Area of low power field of view} = \pi r^2 \text{ or } (3.14)(.880)(.880) = 2.43 \text{ mm}^2$$

$$\text{Average stomata counted from your data table} / 2.43 \text{ mm}^2 = \text{stomata/mm}^2$$

**High power (400x)** field of view diameter = .440 mm (verify this with a micrometer ruler).

$$\text{Area of high power field of view} = \pi r^2 = (3.14)(.220)(.220) = .15 \text{ mm}^2$$

$$\text{Average stomata counted from your data table} / .15 \text{ mm}^2 = \text{stomata/mm}^2$$

Which magnification did you use? \_\_\_\_\_

Show calculations:

8. From your initial observations, ask a question about stomata. Think about the distribution and function of stomata on your leaf. Do you think that all leaves from this plant are the same? Do you think that leaves on different plants have the same distribution? Do all plants have stomata? What plant parts have stomata? Can you control the opening and closing of stomata? How are stomata formed? Do edible vegetables have stomata? How many stomata are on a typical leaf? What other questions do you have?

State a question here:

9. Design a controlled experiment to answer your question:

Design a data table to record your results. Think about how many observations you need to make to see a pattern. Is one sample enough? Think about ways to collaborate with your classmates. Can you find someone who is interested in a similar project?

10. Explain your results and share them with your class.