#### **LEARNING OBJECTIVES**

After completing this exercise, students should be able to:

- Describe the process of and principles behind transpiration.
- Describe how stomata, guard cells, mesophyll cells and vascular tissues interact in the process of transpiration.
- Evaluate how different environmental conditions influence rates of transpiration.
- Compare the results between a control and treatment groups testing principles of transpiration.

Review sections 29.2, 29.4,31.1-31.5 in Mason et al. text.

#### INTRODUCTION

Transpiration is the process whereby water evaporates from a plant and into the air through openings in the leaf called **stomata**. The cohesiveness of the water molecules to one another and adhesiveness to xylem cell walls "pulls" them through the plant body as other water molecules evaporate from cell walls in the leaf **mesophyll**, exit the leaf via the stomata, and enter the atmosphere (Figure 1). This explanation of water movement in plants is called the **Cohesion-Tension Theory**, and it describes how large quantities of water into and through a plant without the aid of any type of pump. Movement of water through the plant by transpiration is driven by differences in **water potential** (given the symbol  $\Psi$ ) between the soil, plant tissues, and air. Water always moves from areas of relatively higher water potential to areas of relatively lower water potential. So, the water potential gradient between the air and the soil provides a driving force for water movement through the plant. Factors that can affect water potential gradients and transpiration include the solutes in water, soil, or plant tissues, atmospheric relative humidity, and temperature.

An important regulator of transpiration is the stomatal complex composed of the opening or stoma (plural = stomata) and the two guard cells that surround it (Figure 2). The word stoma comes from the Greek word for "mouth" and much like the mouth of animals, the stoma opens and closes to allow oxygen to exit and carbon dioxide to enter the leaf. When the stomata are open, water also exits the leaf which generates the tension necessary to draw water from the root and up through the plant. As water is drawn into the plant, it also brings ions from the soil and into the plant to support plant growth. However, opening stomata presents a conflict for the plant Opening the stomata allows water to exit, but if water moves out too rapidly or too much water is lost, the plant can suffer water stress, wilt, and die. However, if stomata close to reduce water loss, internal oxygen levels become too high and carbon dioxide levels become too low resulting in photorespiration. Furthermore, cooling by evaporation decreases when stomata close leading to potentially lethal increases in leaf temperature. On an ecological scale, because transpiration is responsible for moving large amounts of water from the soil and into the atmosphere, it can have a significant influence on the abiotic environment in an area. Thus, how plants regulate and control transpiration is, therefore, a critical physiological feature of plants.

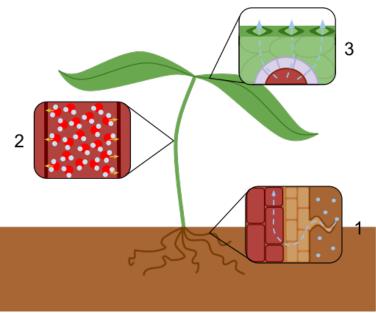


Figure 1. Cohesion-Tension Model of Transpiration. 1-Water molecules enter the roots passively, passing through the cortex, and are guided to xylem tissue by the Casparian strip. A concentration gradient of solutes within the root leads to further passive transport of water into the root. 2-Once the water has entered the xylem, the forces of adhesion and cohesion take effect. Adhesion causes the molecules to adhere to sides of the vessels, and cohesion causes the molecules to adhere to each other, forming a column of water that extends from the roots to the top of the plant. 3-Water from the xylem is let into the spongy mesophyll, where it can come into contact with stomata. When the stomata are open, the cells in this layer are exposed to the outside air, and evaporation occurs. When water molecules evaporate out of the stomata, they create tension in the water column, as the forces of cohesion pull nearby molecules up with them, creating pulling force down the length of the xylem.

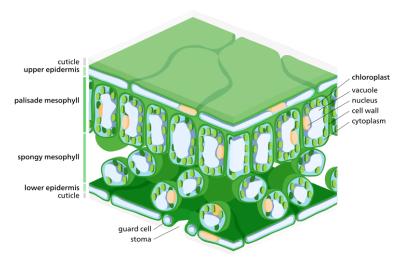


Figure 2. Typical angiosperm leaf tissue structure.

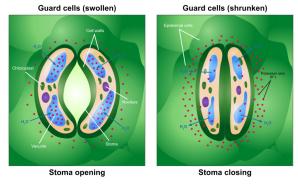


Figure 3. Stomatal complex with guard cells swollen and stoma open (left) and guard cells flaccid and stoma closed (right).

In this laboratory exercise, you will investigate different structural and functional aspects of transpiration. You will observe different components of the plant vascular system and conduct an experimental manipulation of corn ( $Zea\ mays$ ), a  $C_4$  plant, and sunflower ( $Helianthus\ annuus$ ), a  $C_3$  plant. The results of this exploration will demonstrate the fundamental principles of transpiration and the function of plant structures in this important physiological process.

# VISUALIZING STOMATA, GUARD CELLS, & VASCULAR TISSUE Materials

- leaves from two different plant species
- microscope slides
- clear fingernail polish

- clear package sealing tape
- microscope slides of plant tissues
- compound microscope

#### **Procedure**

- 1. Obtain a leaf from a corn plant and a sunflower plant.
- 2. Paint a 1 cm<sup>2</sup> area of clear fingernail polish on the top of the leaf and the undersurface of the leaf, and allow it to dry completely.
- 3. Place a piece of tape on the dried fingernail polish. Gently peel it from the leaf
- 4. Stick the tape on a clean microscope slide.
- 5. Look at the epidermal impression to locate and identify the stomata and guard cells.
- 6. Draw and label the stomata and guard cells. How do they compare between different species?
- 7. Calculate the area of your field of view on your microscope. Count the number of stomata in that area. Repeat for several different regions on your leaf and record your data.
- 8. Calculate the average number of stomata per cm<sup>2</sup> on the top and bottom of the leaves for both species. How do they compare

# For Further Study

- 1. Obtain a box of microscope slides and review the arrangement of vascular tissues in the herbaceous monocot and dicot stem, root, and leaves.
- 2. Compare the vascular tissues of dicot and gymnosperm wood.

### MEASURING TRANSPIRATION BETWEEN A MONOCOT AND DICOT

#### Materials:

- 6 corn plants\*
- 6 sunflower plants\*
- plastic bags and twist-ties

- balance
- fluorescent lamps, incandescent lamps, fans, etc.

## **Procedure**

- 1. Obtain your experimental plants and take them to your bench.
- 2. Place the potted plant in a plastic bag and carefully seal the bag around the stem using the twist-ties.
- 3. Weigh each plant and record its initial weight in your notebook.
- 4. Place your experimental plants in the appropriate experimental conditions you are testing. See Table 1 for examples.
- 5. After 15 minutes, weigh each plant again.
- 6. Continue taking measurements until your plants have been in their experimental conditions for 90 minutes.
- 7. Convert the change in weight to volume of water transpired. (Remember 1g  $H_2O = 1$  ml.)
- 8. Next calculate the cumulative % change in plant weight.
  - Cumulative % Change = ((Weight at  $t_0$  Weight at time  $t_n$ )/weight at  $t_0$ )
- 9. Plot the volume of water transpired (y-axis) versus time (x-axis).
- 10. Plot the cumulative % change in plant weight (y-axis) versus time (x-axis).

# For Further Study

- 1. After the last transpiration measurement, harvest the aboveground portions of the plant. Remove all of the leaves from the plant and weigh the stem and leaves separately. This is the fresh weight.
- 2. Place the leaves in a brown paper sack and allow them to dry in a lab oven set on low temperature. After leaves have dried thoroughly, weigh them again. This is the **dry weight**.
- 3. Calculate and plot the volume of water transpired/g fresh weight and volume of water transpired/g dry weight. How do they compare? How do the values compare between species?

<sup>\*</sup> Each group will work with either corn or sunflower, but will be paired with a group using the other species. Plants were watered prior to lab so the soil is saturated.

## **ASSIGNMENT BEFORE LAB:**

In your lab notebook,

1. Prepare tables for collecting data from your transpiration experiment. You will need to collect data from your 6 plants and the 6 plants of the group you will be paired with for this study. Class data will be compiled and shared among all groups.

2. Reproduce the table below in your notebook and predict how each of the listed environmental variables will affect transpiration? Explain the rationale for your predictions.

T     4 D	r 1.cc	1 11.1
Lable 1. Predicted effects	ot ditterent environment:	al conditions on transpiration.
rabio ii realetea effects		ar corrattions on transpiration.

Condition	Predicted overall	Predicted effect	Explanation.
	effect (increase or	on C <sub>3</sub> vs. C <sub>4</sub> plant	
	decrease in		
	transpiration)		
Control			
Fan			
Mist			
Darkness			
Supplemental			
Light			

# Image Credits

Transpiration\_Overview.svg By Laurel Jules (Own work) [CC BY-SA 3.0 (https://creativecommons.org/licenses/by-sa/3.0)], via Wikimedia Commons. Available at https://commons.wikimedia.org/wiki/File:Transpiration\_Overview.svg

Leaf Tissue Structure By User:Zephyris, modified by Kelvinsong. (File:Leaf Tissue Structure.svg) [CC BY-SA 3.0 (https://creativecommons.org/licenses/by-sa/3.0)], via Wikimedia Commons Available at: https://commons.wikimedia.org/wiki/File%3ALeaf\_tissue\_structure\_flat.svg

Opening and Closing of Stoma.svg By Ali Zifan [CC BY-SA 4.0 (https://creativecommons.org/licenses/by-sa/4.0)], via Wikimedia Commons. Available at https://commons.wikimedia.org/wiki/File:Opening\_and\_Closing\_of\_Stoma.svg

