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Rate of Transpiration

AP* Biology Big Idea 4, Investigation 11

An Advanced Inquiry Lab

Introduction

All living things acquire nutrients, ions, and water from the environment. Plants absorb these materials from the surrounding soil and air via the processes of osmosis, diffusion, and active transport. The water, nutrients, and ions are then transported throughout the plant within xylem because of differences in water potential.

Concepts

- Adhesion
- Biotic vs. abiotic factors
- Cohesion
- Osmotic potential
- Stomata
- Transpiration
- Turgor potential
- Water potential

Background

Water must follow the laws of thermodynamics. Consequently, water always moves from regions of high energy to regions of lower energy. In a plant this means that water flows from regions of high water potential to regions of lower water potential. This occurs through the processes of osmosis, root pressure, and adhesion and cohesion of water molecules. In plants, water potential is equal to the sum of the osmotic potential (ψ_s) and the turgor or pressure potential (ψ_p).

Osmotic potential is a measure of the amount of solutes (dissolved minerals and other nutrients) in the water within the plant. The root cells expend energy to actively transport dissolved minerals into the root. An increased solute concentration in the roots causes a lower amount of free energy and therefore a negative water potential in the root tips. Water then flows by osmosis from the region of high energy in the soil to the region of lower energy in between the root cells. Osmotic potential is always negative in a plant.

Turgor potential occurs when water molecules enter a cell and apply pressure to the cell walls. Living plant cells have positive turgor potential. The cells in wilted leaves have zero turgor potential. Specialized water transport cells called xylem have a negative turgor pressure because water is removed from xylem by the adjacent cells due to osmosis. Plants require a consistent supply of water around their roots because they constantly lose water through their leaves via transpiration. *Transpiration* is the loss of water by evaporation from the leaves and is the main method for pulling water from the roots to the leaves.

Transpiration begins with evaporation of water through the *stomata* (singular: stoma or stomate). Stomata are tiny openings (pores) used for the absorption of carbon dioxide (CO_2) for photosynthesis and oxygen (O_2) for cell respiration (see Figure 1). Thousands of stomata occur on the underside of a typical dicot or on the upper surface of a plant whose leaves float on water. Each stoma is formed by a pair of specialized cells known as *guard cells* that are responsible for regulating the size of the pore's opening. By adjusting the size of the opening, the guard cells control the rate of CO_2 and O_2 uptake and the loss of water by the leaf. In this way, by regulating the diffusion of CO_2 into the cells, the guard cells also control the rate of photosynthesis in the leaf. The guard cells swell when they are full of water, opening the stoma into air spaces that surround the middle layer of leaf cells. This middle layer of cells is called the mesophyll (*meso*=middle, *phyll*=leaf). The mesophyll cells are covered with a thin layer of water from the xylem. The water coating the cells evaporates due to the lower water potential in the outside air. New water molecules then move onto the mesophyll cells by osmosis from the xylem.

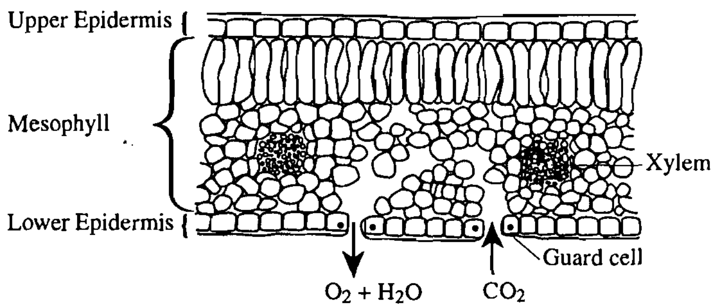

Figure 1.

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As each water molecule moves onto a mesophyll cell, it exerts a pull on the column of water molecules in the xylem, from the leaves to the roots (see Figure 2). This transpirational pull is caused by the cohesion of water molecules to one another due to hydrogen bond formation, and by the adhesion of water molecules to the walls of the xylem cells. The upward transpirational pull on the fluid in the xylem causes negative pressure to form in the xylem, pulling the xylem walls inward and creating decreased water potential inside the xylem. This decrease in water potential is transmitted all the way from the leaves to the roots. Water then moves inward from areas of higher water potential in the soil, through the root hairs, and into the xylem.

If the moisture content in the mesophyll layer of the leaf equals or is less than the moisture level of the outside air, the guard cells will lose their water, and the cells will become flaccid and close. Many environmental conditions influence the opening and closing of the stomata and thus affect the rate of both transpiration and photosynthesis. Temperature, light intensity, air currents, humidity, and the nature of the plant all influence the guard cells to open or close.

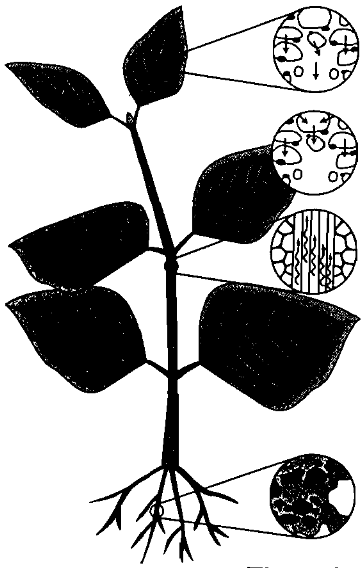

Figure 2.

Experiment Overview

In the *Baseline Activity,* the stomata, which regulate transpiration, will be observed, counted and quantified. A method will be developed to determine the approximate leaf area and therefore the approximate number of stomata for each plant. The study of stomata and leaf area provide a foundation for the development of an open-inquiry, student-designed experiment—see the *Opportunities for Inquiry* section. Biotic, abiotic, and altering experimental techniques are all variables that may affect the rate of transpiration. The rate of transpiration of a control will then be mathematically compared to that of the treatment.

Pre-Lab Questions

1. Very few plants can tolerate salty conditions such as found on the salt flats or along a sidewalk that has been treated with de-icer over the winter. Why do salts affect plants this way? Create a well-labeled diagram to explain.
2. In the *Background* section the location of stomata in a land plant and a plant with leaves that float on water was discussed. What about plants that stay submerged in water? Do they have stomata at all and if so where are they located on the plant?

Materials

Calculator	Microscope slides
Clear cellophane tape (clear package sealing tape)	Plant leaves
Clear fingernail polish	Ruler, clear
Microscope	Scissors

Safety Precautions

Nail polish is toxic by ingestion and inhalation. Avoid eye contact. Wash hands thoroughly with soap and water before leaving the laboratory. Please follow all laboratory safety guidelines.

Baseline Activity

1. Paint a thick patch of clear nail polish on the leaf surface being studied. Make the patch at least one square centimeter.
2. Allow the nail polish to dry completely.
3. Place a piece of clear cellophane (packing) tape onto the dried nail polish patch.
4. Gently peel the nail polish patch from the leaf by pulling on a corner of the tape and "peeling" the fingernail polish off the leaf.

5. Tape the peeled impression to a clean microscope slide.
6. Count all the stomata in one microscopic field and repeat for at least four leaf areas.
7. Use a clear ruler to determine the size of one field of view under the microscope.
8. Calculate the number of stomata per square millimeter.
9. Pool the class data and determine the average number of stomata per square millimeter of the plant species being tested. The total number of stomata varies by species. Since transpiration occurs via the stomata, this value is an important consideration when determining the transpiration rate.
10. The rate of transpiration for a plant is generally reported as volume of water (in milliliters) or the mass of water (in grams) per square meter of leaf area. Develop a procedure to determine the total leaf area of a plant.
    - *a.* The plants may not be destroyed until after testing is complete. However, the remainder of the plant used for the stomata counting activity may be used to help determine the leaf area.
    - *b.* Each group should develop at least one method to estimate the total leaf area of the plants.
    - *c.* Decide, as a class, the procedure that will be used to determine leaf area for the remainder of this laboratory.
    - *d.* The total leaf area and the approximate number of stomata on each plant will need to be calculated as part of the data analysis section of the inquiry portion of this lab.

Opportunities for Inquiry

1. Consider the following questions while reflecting upon your knowledge about transpiration in a flowering dicot.
    - *a.* How does the weather and environment affect transpiration?
    - *b.* How does the number of stomata per square meter of leaf area affect the transpiration rate?
    - *c.* Do different waters, liquids, or ions affect transpiration rate?
    - *d.* Does one testing method have a lower experimental error than another?
    - *e.* How does coating the leaves or otherwise compromising the plant affect transpiration rate?
    - *f.* Are there ways to increase transpiration rate?
2. Plan, discuss, execute, evaluate, and justify an experiment to test a question regarding transpiration.
    - *a.* Decide upon one question that your group would like to explore.
    - *b.* Develop a testable hypothesis.
    - *c.* Discuss and design a controlled procedure to test the hypothesis.
    - *d.* List any safety concerns and the precautions that will be implemented to keep yourself, your classmates, and your instructor safe during the experimental phase of this laboratory.
    - *e.* Determine what and how you will collect and record the raw data.
    - *f.* How will you analyze the raw data to test your hypothesis?
    - *g.* Share your hypothesis, safety precautions, procedure, data tables, and proposed analysis with your instructor prior to beginning the experiment.
    - *h.* Once the experiment and analysis are complete, evaluate your hypothesis and justify why or why not the hypothesis was supported by your data.
    - *i.* Present and defend your findings to the class.
    - *j.* Make suggestions for a new or revised experiment to modify or retest your hypothesis.

Option 2. Whole Plant Method

Materials

Balance, 0.001-g precision

Small potted plants, 2

Bags, plastic 11″ × 22″, 2

Rubber bands, 2

Water, tap

Safety Precautions

Although the materials in this lab activity are nonhazardous, follow normal safety precautions. Wash hands thoroughly with soap and water before leaving the laboratory.

Procedure

1. Saturate the plant with water the day before starting the inquiry activity.
2. Remove a plant from the pot. Keep the roots and as much soil as possible.
3. Place the root ball and soil into the plastic bag and secure with a rubber band (see Figure 4).
4. Remove all flowers and buds.
5. Determine the mass of each plant over the course of a week as the plant is exposed to the experimental conditions.

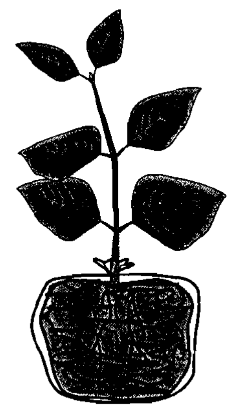

Figure 4.