# Which Way To Grow: Exploring Tropic Responses of Plants

#### SYNOPSIS FOR CORE EXPERIMENT

Students will assess the influence of seed orientation on the direction of root growth in corn seeds.

#### APPROPRIATE BIOLOGY LEVEL

Introductory or advanced

### **Directions for Teachers**

*Note to Teachers:* Information below is given for the Core Experiment. Additional information needed for each variation of the Core Experiment may be found beginning on page 176. For a specific variation, check the At-A-Glance Map.

#### GETTING READY

See sidebars for additional information regarding preparation of the lab.

#### **OBJECTIVES FOR CORE EXPERIMENT**

At the end of this lab, students will be able to:

- Explain how gravity is thought to cause the geotropic response in a corn plant.
- Measure angular differences in root growth and interpret these data with statistical analysis.

#### MATERIALS NEEDED

You will need the following for a class of 24:

- 1 shallow, 24 x 28-cm glass pan
- I L distilled water
- 144 corn seeds

You will need the following for each group of two students in a class of 24:

- 1 package of mounting putty or 12 straight pins
- 12 presoaked corn seeds
- 4 pieces of 10-cm filter paper or 10 x 10-cm chromatography paper
- I plastic food storage box with lid
- I protractor
- 1 pencil

#### SAFETY PROCEDURE

Handle fungicide-treated seeds with forceps or gloved hands. (See Teaching Tips.)

) Use glues or sealants in a hood or ventilated area.

### DIRECTIONS FOR SETTING UP THE LAB

One day before

Place seeds in a shallow pan of water 2.5 cm deep overnight for 12 hours.

#### **TEACHER PARTNERS**

Anthony Umelo and Navarro Bharat Anacostia Senior High School 16th and R Streets, SE Washington, DC 20020



#### LENGTH OF LAB

A suggested time allotment follows:

- Day 1 (45 minutes)
- Observe sample root orientations, then discuss and form hypothesis.
- Day 2 (20 minutes)
- Attach seeds to paper and arrange them in germination chamber.
- Day 3 or 4 (45 minutes)
- Collect and share data. Introduce analysis to be completed outside of class or on another day when individual variations are designed.

#### PREPARATION TIME REQUIRED

- 10 minutes
- Gather supplies.
- 12 hours
- Soak the seeds.

Which Way To Grow

#### TEACHING TIPS

- Corn is not bred for a gravitropic response, so one may expect a large variation in response even in the same seed lot of a variety (Matthews & Zobel, 1995).
   Compensate by pooling data to create a large sample of 50 or more plants.
- The scientific name of corn is *Zea mays*.
- It is important to provide a smooth surface for the growth of roots so that they do not respond to obstacles, such as wrinkles in the paper or small moisture differences, rather than gravity.
- Presoaking the seeds makes it easy to pin them through the endosperm. Alternatively, you can use mounting putty or glue to affix them to the paper while they are dry, and then soak them overnight before orienting them in the germination boxes. Glues that work well include Amazing Goop<sup>TM</sup>, Duro<sup>®</sup> Super Glue, RoSS<sup>TM</sup> Rubber Cement, and Dap<sup>TM</sup> Bathtub 100% Silicone Sealant. Check the labels of these products for hazards and any safety considerations. These adhesives hold tight when wet and do not inhibit germination when applied to dried seeds. Very sweet corn or "Supersweet" corn varieties may break the bond with an adhesive as the seed hydrates. Most seeds will hold fast.
- If you do not have equipment to project the results of a single sample for introducing the experiment, you may photocopy the germination chamber and make a transparency of it. You also can scan the results of a petri dish and display them on the computer monitor or print a transparency. Alternatively, you could pass the chamber(s) from student to student.
- Sweet corn seeds shrivel. It is difficult to identify or orient the embryo in some varieties. If you presoak the corn, it will plump and the embryo will be easier to see. See Figure 1.
- Sweet corn is frequently treated with fungicide. The fungicide will reduce problems with contamination, but requires that you take extra care in handling

#### TEACHER BACKGROUND

#### Content Information

Corn, like almost all plants, grows vertically with the shoot upward and the root downward. Plants, as germinating embryos, are responsive to up and down. If they were not, their shoots would not reach the light needed for photosynthesis, and their roots would not reach water and nutrients essential for growth and metabolism. The plant's response to up and down is called gravitropism (formerly called geotropism).

Plant signal perception is an evolutionary adaptation to the immobile nature of plants, giving them an ability to respond to environmental stimuli. Perception of gravity is thought to occur by sedimentation of amyloplasts in specialized cells called statocysts located in the root cap (Konings, 1995). The gravity stimulated curvature of a root is controlled by the movement of auxin that inhibits root growth. Lateral roots accumulate a higher concentration of auxin on their lower side. Elongation of cells on the lower side is inhibited relative to the cells on the upper side, and the root curves downward. High auxin concentrations stimulate stem growth. The connection between auxin concentration and amyloplast sedimentation is not entirely clear, but the sedimentation may somehow cause a movement of calcium ions that influence the transport of auxin.

#### Pedagogical Information

The following is a chart of some concepts related to this lab and some student misconceptions of these concepts.

<ul> <li>Correct Concept</li> <li>A seed is composed of differen- tiated tissue. Only the embryo develops into a new plant.</li> </ul>	<ul><li>Misconception</li><li>The entire seed grows into a plant.</li></ul>
• Cotyledons are immature leaves of the plant embryo. The coty- ledons of dicots, such as bean, no longer contain endosperm but still serve as food reserves. In monocots such as corn, en- dosperm is still present but sepa- rate from the cotyledons.	<ul> <li>Both monocot and dicot cotyle- dons are immature leaves of the embryo that contain endosperm.</li> </ul>
• Seeds may be germinated in vitro and embryos grown on chemically defined media.	• Seeds must be planted in soil in order to grow.
<ul> <li>Specific plant hormones pro- mote differential growth.</li> </ul>	• Fertilizers are the only chemicals that promote seed growth.

- Seeds are living.
- Seeds are nonliving.

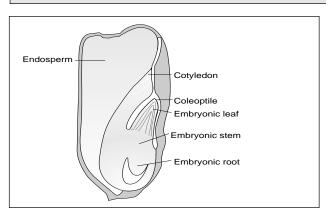


Figure 1. The structure of a corn seed.

#### INSTRUCTIONAL PROCEDURES FOR THE CORE EXPERIMENT

#### Introduction

Present five or six kinds of seeds that might be planted in gardens in your area. Tell a story about your father or mother always insisting that you plant seeds right side up. Ask students if you were just being teased about taking more time to plant those seeds, or if your parent was serious. Ask them if they can tell which is the right side on the seed. For most seeds this is very difficult. They are unlikely to be able to tell.

Four or five days before introducing the lab, affix the corn seeds on the filter paper with mounting putty in the pattern shown in Figure 2. Soak and germinate the seeds.

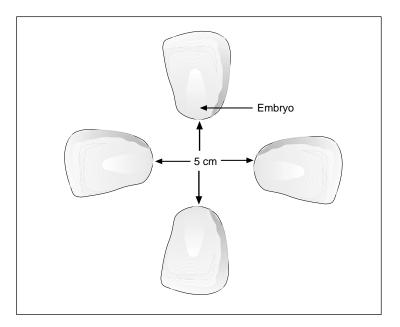


Figure 2. Seed positioning pattern for introducing the lab.

As the students make observations of the specimens, the teacher can prompt a discussion with the following questions:

- Can you tell which is right side up for these corn seeds?
- Would it make a difference how they were oriented when they were planted?
- In which direction is each root and shoot growing?
- Which way do you think was up when these plants started to grow?
- Discuss and demonstrate regular measurement techniques used in the Core Experiment.

#### HYPOTHESIS GENERATION

The following discussion and activities are designed to elicit questions that students can transform into hypotheses. Questions that may be elicited include the following:

- What would happen to a seedling that did not grow the way these seedlings are growing?
- What could influence how a seedling grows?
- What is the cue to send the shoot toward the surface if a seed is planted deeper in the soil than light penetrates?
- What would happen if these seedlings were germinated in space?



#### TEACHING TIPS

the seeds. You can recognize the fungicide by its bright pink color.

- Do not reorient the seeds once the germination chamber has been set up. A root may respond by changing the direction of growth when stimulated by gravity for only 2 minutes (Gonick, 1997) or in as little as 27 seconds (Perbal & Ecole, 1995).
- Normally, embryo and root or shoot angles are measured from the vertical. Straight up is considered 0° and straight down is considered 180°.
- Do not incubate the seeds in total darkness. The gravity response is reported to be stronger in roots exposed to white light (Kelly & Leopold, 1992).

Which Way To Grow

### TEACHER'S NOTES

#### Sample Hypotheses

- If gravity is the cue that orients root growth, corn seeds planted at various angles will all have roots growing in the same direction.
- If a temperature differential is the cue that orients root growth, seeds heated from below and cooled from above will not produce roots oriented downward.

On the following pages are a sample hypothesis, procedure, and data analysis set with interpretation that students might develop for the Core Experiment. It is followed by a related test question and answer for teacher evaluation. This example has been included as a potential outcome of the activity and should not be given to the students. Students should develop their own hypotheses and procedures. Make sure they understand that there is not just one correct hypothesis, procedure, or data set. The Variations of the Core Experiment will give each team of students the opportunity to expand on the Core Hypothesis. Additional test questions are found on page 175.

#### Question

Is there a difference in angle of root growth and the displacement of the embryo if corn seeds are oriented with the embryo tilted from its horizontal axis?

#### Hypothesis

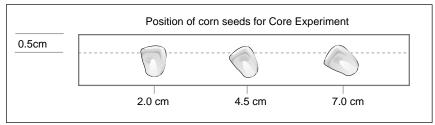
If corn seeds are oriented with the embryo tilted from its horizontal axis, then the difference between angle of root growth and the displacement of the embryo from the vertical position will increase with reference to the angle of the force of gravity.

#### Rationale

Roots of germinating plants must anchor the plant and reach water and nutrients even if they are not hand planted by humans. A large difference between orientation and final position indicates a great orientation to gravity by the root.

#### Procedure

- 1. Remove 12 presoaked corn seeds from the pan of seeds soaked overnight. Place on a paper towel.
- 2. Draw 3 small dots with a pencil on each of 4 pieces of filter paper. Position the dots 0.5 cm from the top of the paper at 2.0, 4.5, and 7.0 cm from the left edge. See Figure 3.
- 3. Mount the presoaked corn seed on each dot of the paper as shown in Figure 3. Do not allow the seeds to dry while you work with them.



**Figure 3.** Position of corn seeds for the Core Experiment. Seeds may be affixed with mounting putty or straight pins.

- 4. Wet the paper completely.
- 5. Create a germination chamber by placing a little mound of modeling clay at 8 evenly spaced locations on the bottom of a plastic box.
- 6. Use the straight edge of your protractor to make a groove parallel to the far side of the box through each clay mound.

- 7. Stand each piece of filter paper in a pair of the grooves.
- 8. Add water to the bottom of the dish to 0.5-cm depth. Be certain that the entire bottom edge of each piece of paper is in the water.
- 9. Seal the lid on the germination chamber and incubate the seeds at room temperature.
- 10. When the root is 10 to 15 mm long, measure the angle of embryo displacement and the angle of root growth.
  - a. Draw a straight line through the long axis of the embryo.
  - b. Draw a straight line from the hypocotyl to the tip of the root.
  - c. Use a protractor to measure the angles in degrees from the vertical line as indicated in Figure 4 and record your results in a table.

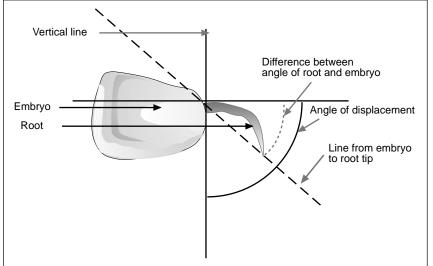


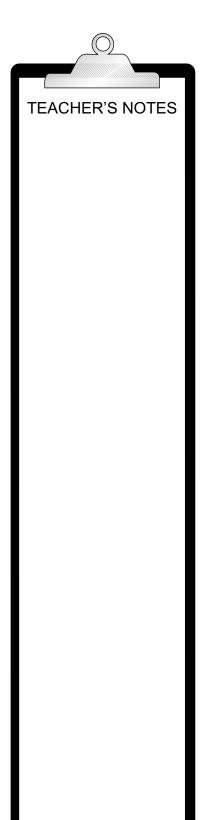
Figure 4. Angles to measure for determining root response to gravity.

11. Pool class data for analysis.

# DATA ANALYSIS AND INTERPRETATION Sample Data

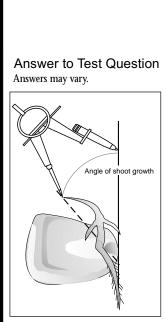
Table 1. Effect of corn embryo angle on the angle of root growth.

Student team	Angle of displacement	Difference between embryo and root growth angles
1-Plant 1	43	50
1-Plant 2	77	37
1-Plant 3	120	82
1-Plant 4	33	7
2-Plant 1	93	33
2-Plant 2	90	61
2-Plant 3	93	91
2-Plant 4	105	87



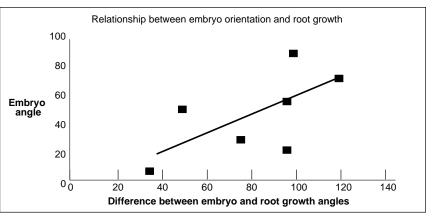


The data support the hypothesis that root response to gravity results in a stronger root curvature when the embryo is tilted more from the vertical.



**Figure 5.** Instructions for measuring shoot angle of corn seedlings.

Draw a straight line from the tip of the shoot to the vertical line on which the seed was glued. Use a protractor to measure the angle between the line you have just drawn and the vertical line.



**Graph A.** Comparison of corn embryo orientation and final root growth angle after 3 days incubation in the dark at room temperature. Angles were measured in degrees. The line is a least squares regression line.

#### TEST QUESTION

Suppose you are interested in testing the gravitropic response of shoots. How would you measure that response? Draw a figure and write an explanation to clarify your technique.

#### STUDENT DESIGN OF THE NEXT EXPERIMENT

After students have collected and analyzed these data from their experiments and shared results and conclusions with the class, encourage them to brainstorm ideas for experiments they could do next. They should think of questions that occurred to them as they conducted their first experiments. Ask them what quantifiable experiments could be done based on observations they have made.

Have students return to their experimental lab groups to share ideas before writing their proposals. Questions students may suggest include the following:

- What other environmental factors might influence gravitropic responses? How might you independently measure the effects of these factors? Why do these factors influence gravitropic responses?
- What other environmental stimuli might plants respond to and why? How would you test these responses independent of one another?

#### SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

These are possible ways to modify this specific activity for students who have special needs, if they have not already developed their own adaptations. General suggestions for modification of activities for students with disabilities are found in the AAAS *Barrier-Free in Brief* publications. Refer to p. 15 of the introduction of this book for information on ordering FREE copies of these publications. Some of these booklets have addresses of agencies that can provide information about obtaining assistive technology, such as Assistive Listening Devices (ALDs); light probes; and talking thermometers, calculators, and clocks.

#### Blind or Visually Impaired

Materials that are needed by students who are visually impaired include a braille protractor, braille ruler, braille paper, braille graph paper, stylus, wax pencil, rubber mat or base for making raised-line drawings, tracing wheel, blunt tip tools for drawings, talking thermometer, talking watch or timer, and rubber gloves for handling fungicide-treated seeds.

Which Way To Grow

- Provide the blind student with a copy of the lab experiment in braille with graphs on braille graph paper and raised-line drawings. Plant experiments often can be performed by students who are blind.
- Provide interpretation of the direction "mount seed as shown in Figure 3."
- Explain where the *hypocotyl* is located in Step 10. Locate supplies within easy access of students who are blind to allow them to do this step unaided.

#### Deaf or Hard-of-Hearing

No special modifications are needed for this experiment.

#### Gifted

- Have gifted students design experiments to propose possible explanations for the gravitropic responses on the cellular level. Challenge them with questions such as, "Are gravitropic responses due to changes in the distribution of cells or hormones within plant tissues?"
- Encourage students to undertake studies to determine what part environmental factors and/or genetic factors play in the plants' responses to gravitropism. A combination of library research and discussions with scientists in nearby universities or research facilities will help in this regard.

#### Mobility Impaired

The general directions for performing this activity can be followed by students in wheelchairs. See "Allelopathy" for details.

- Have manually impaired students act as observers. This experiment requires fine motor skills to handle the seeds and seedlings.
- Provide access to a computer if manually impaired students have computer skills to compile data and do graphing.

#### ADDITIONAL TEST QUESTIONS

Test questions for the Core Experiment also may include the following:

- 1. If an embryo is at an angle of 90° and the root grew downward at 60°, how far from the vertical position would the root be growing? How does this root's orientation to the vertical compare with your observations?
- 2. Why was it desirable to pool class data to evaluate the effect of gravity on the angle of root growth?

#### REFERENCES AND SUGGESTED READINGS

- Barclay, G.F. & Clifford, P.E. (1991). Using light microscopy to study geotropism. *The American Biology Teacher*, *53*(5), 285-286.
- Bjorkman, T. & Cleland, R.E. (1991). The role of extracellular free calcium gradients in gravitropic signaling in maize roots. *Planta*, *185*(3), 379-384.
- Clifford, P.E. & Oxlade, E.L. (1991). Using dandelion flower stalks for gravitropic studies. *The American Biology Teacher*, *53*(5), 290-293.

Gonick, L. (1997). Starch wars. Discover, 18(8), 32-33.

- Guisinger, M.M., Miller, A. & Kiss, J.Z. The Response to Gravity Is Correlated to the Amount of Starch in *Arabidopsis* hypocotyls. [Online]. Available *http://www.indstate.edu/mwaspp/1996\_meeting/suisinge.html*, July 1997.
- Kelly, M.O. & Leopold, A.C. (1992). Light regulation of the growth response in corn root gravitropism. *Plant Physiology, 98*(3), 835-839.
- Kim, S.Y. & Mulkey, T.J. (1994). Auxin movement during root gravitropism. *Bioscience, 20*(1), 7-10.

Konings, H. (1995). Gravitropism of roots: An evaluation of progress during the last three decades. *Acta Botanica Neerlandica, 44*(3), 195-223.

#### Answers to Additional Test Questions

**TEACHER'S NOTES** 

- 1. This root would be only  $30^{\circ}$  from straight down. Our average angle between the root and the vertical line is about  $24^{\circ}$ , so these values are similar.
- 2. Corn plants vary widely in their response to gravity. To detect a pattern, it is necessary to have a large sample size.



#### Answers to Questions and Analysis on Student Page

- 1. The graph should be similar to Graph A. The combined class data show that the greater the angle of the embryo, the more the root curved.
- 2. These data support the hypothesis. The roots do compensate for greater original disorientation by growing with a greater curvature, finally growing nearly straight down.
- 3. If one wanted to create a variety of corn with as great a tropic response as the most strongly responding plant, one would clone it. Alternatively, one could selfpollinate the mature plant, or at least cross pollinate two plants with strong responses. Cloning would be faster and probably less expensive.
- 4. Other stimuli that could cause the roots to change their angle of growth might be touch, such as that presented by coarse grains in the soil, and a directional supply of water.

- Laferriere, J.E. (1993). Competitive gravitropic & phototropic stimuli in Coleus. The American Biology Teacher, 55(6), 370-371.
- Lee, J.S., Mulkey, T.J. & Evans, M.L. (1983). Reversible loss of gravitropic sensitivity in maize roots after tip application of calcium chelators. Science, 220(4604), 1375-1376.
- Matthews, L. & Zobel, R. (1995). Gravitropic responses of five maize hybrid roots to temperature. Journal of American Institute of Aeronautics and Astronautics, 1-6.
- National Corn Growers Association. (1997). Corn fagtory: Frequently Asked Questions-FAQ. http://www.ncga.com/04faqtor/page041.html.
- Perbal, G. & Ecole, D.D. (1995). Sensitivity to gravistimulus of the lentil root. [Online]. Available http://www.publinet.it/elgra//perbal.html, June 1995.
- Smith, M. (1991). Plant growth-responses to touch-literally a 'hands-on' exercise! The American Biology Teacher, 53(2), 111-114.
- Sokal, R.R. & Rohlf, F.J. (1995) Biometry. The Principles and Practice of Statistics in Biological Research (2nd ed.). New York: W.H. Freeman and Company.
- Takahashi, H. (1994). Hydrotropism and its interaction with gravitropism in roots. Plant and Soil, 165, 301-308.
- Van der Pijl, L. (1972). Principles of Dispersal in Higher Plants (2nd ed.). Springer-Verlag: New York.

Zzzzz. How roots respond to gravity. Scientific American, 254, 112-119.

### VARIATIONS ON THE CORE EXPERIMENT

After completing the Core Experiment, students should use the results to develop a variation on that experiment. The following directions are meant only as a guide for the teacher. They suggest possible hypotheses students may develop and data that may result.

*Note to Teachers:* Only information that is unique to each Variation of the Core Experiment is found in this section. Unless otherwise noted, teacher information not listed for each variation is the same as that found in the Core Experiment. Materials listed in this section are needed in addition to the materials listed for the Core Experiment.

#### VARIATION 1

#### The Effect of Temperature on the Gravitropic Response of Corn

#### **SYNOPSIS**

Students will compare the root angle of the same variety of corn under two or more temperatures.

#### ADDITIONAL MATERIALS NEEDED

You will need the following for a class of 24 students:

Two or more temperature controlled environments, such as incubators or water  $\bigcirc$ baths.

#### HYPOTHESIS GENERATION

#### Question

What is the effect of increased temperature on the root growth angle of corn seedlings?

#### **TEACHING TIPS**

- Most classrooms are not controlled at the same temperature throughout the day. You could use an unheated basement, where the temperature is fairly constant over a period of several days, and a deep pan of water with an inexpensive aquarium heater to provide two temperature regimes if you do not have incubators or water baths.
- Matthews and Zobel (1995) found variable responses of corn roots to different temperatures. The roots grew most directly downward at 25°C.



#### Sample Hypothesis

If the temperature is increased, then the root growth angle of corn seedlings will increase.

#### Rationale

It would be adaptive for roots to grow downward more directly if it is hot, because high temperatures are often accompanied by drying conditions.

#### Sample Experimental Procedure

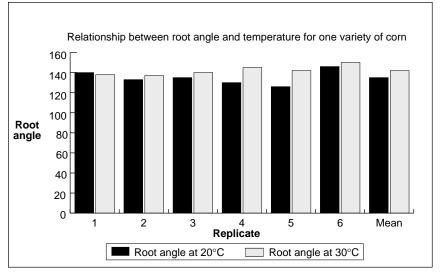
Repeat the Core Experiment placing some germination chambers in each of two or more constant temperature environments.

#### DATA ANALYSIS AND INTERPRETATION

#### Sample Data

**Table 2.** Corn root growth angles measured after incubation in 2 different constant temperature environments.

Replicate	Root angle at 20°C	Root angle at 30°C
1	140	138
2	133	137
3	135	140
4	130	145
5	126	142
6	146	150
Mean	135	142



Graph B. Root angle and temperature relationship.



TEACHER'S NOTES

An appropriate analysis for data collected in this design would be a Wilcoxon Two-Sample Test (Sokal & Rohlf, 1995). To perform the statistical test:

1. Convert measurements to ordered ranks and sum the ranks:

- a. Keep track of 20°C and 30°C values as you order them together from low to high. It is often easiest to do this if you use different colors or two columns to identify these data sets.
- b. Assign ranks to the ordered values. If two observations have the same value, assign them the average of their ranks. For example, both sets of data have the value 140. The ranks for this value are 7 and 8, so each value is assigned the average of (7 + 8) or 7.5.
- c. Sum the ranks of the smaller sample if the samples are of unequal size. Call this smaller sample  $n_2$ , and let  $n_2$  be the number of observations in that sample. The sum of the ranks of the small sample is  $\sum \mathbf{R}_2$ . Call the other sample  $n_1$ , and let  $n_1$  be the number of observations in that sample. The sum of the ranks of that sample is  $\sum \mathbf{R}_1$ . Sum the ranks of either sample if they are of the same size. d. Table 3 shows these steps produce:

Table 3. Result of Wilcoxon Two-Sample Test.

	20° Root angle		30' Root	angle
	Ordered value	Rank	Ordered value	Rank
	126	1		
	130	2		
	133	3		
	135	4		
			137	5
			138	6
	140	7.5	140	7.5
			142	9
			145	10
	146	11		
			150	12
Sum of the ranks $(\Sigma \mathbf{R})$		28.5		49.5

2. Compute: Compute the Wilcoxon statistic by substituting your values in the formula

 $C = n_1 n_2 + [n_2(n_1 + 1)/2] - \sum R_2$ Also calculate C by the formula  $C' = n_1 n_2 - C$ Here these computations produce C = 6(6) + [6(6 + 1)/2] - 28.5 = 28.5

and C' = 6(6) - 28.5 = 7.5

3. Compare: Use the larger of the two values, C or C', as the test value. Compare your value with the tabled value.

<b>n</b> <sub>1</sub>	$\mathbf{n}_2$	Comparison value
3	3	9
4	3	12
4	4	15
5	2	10
5	3	14
5	4	18
5	5	21
6	2	12
6	3	16
6	4	21
6	5	25
6	6	29

Table 4. Critical values at 5% for Wilcoxon Two-Sample Test.

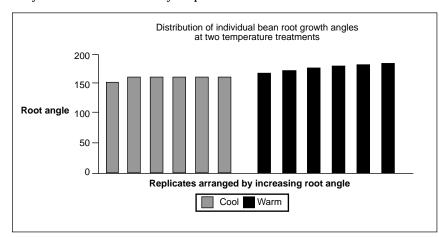
These steps produce:

 $C_{calculated} < C_{tabled}$  or 28.5 < 29

4. Conclude: If your value is less than the tabled value, the apparent differences are most likely the result of chance alone and do not represent significant differences in these data sets. If your value is equal to or greater than the tabled value, the differences in your data sets are not due to chance alone.

#### TEST QUESTION

The graph below displays data collected by another group of students studying the gravitropic response of a single variety of beans to two different temperatures. Does temperature alter the response of the roots of this variety of beans? Is it necessary to analyze these results statistically? Explain.



**Graph C**. Influence of two constant temperature regimes on the gravitropic response of bean roots.

## SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

Blind or Visually Impaired

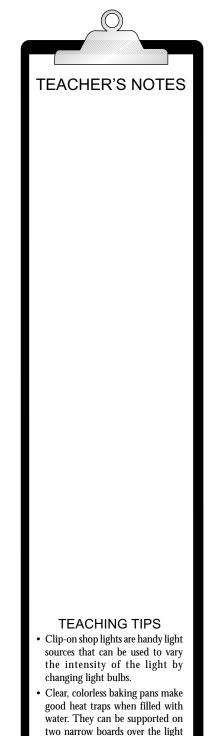
- Provide a talking thermometer for the student who is blind.
- Consider that it may be more difficult for some students to read columns of



Interpretation Statistical analysis reveals that the responses of the roots to gravity do not differ at the two temperatures tested. The variation within a population means that one should probably use a larger sample. However, at this point it must be concluded that the hypothesis is not supported by these data.

#### Answer to Test Question

These students found that the bean roots grew more vertically at the warm temperature. It is not necessary to analyze these data statistically because, although the curvature of all the roots is similar, the cool temperature curvature is always less than the warm temperature curvature and the variation within the samples is similar.



- source · A glass bread pan covered with plastic wrap makes a good clear, colorless germination chamber.
- · Roots kept in the dark have a far smaller (10°) gravitropic curvature in 2 hours than do roots exposed to white light (60°) (Kelly & Leopold, 1992). The response of root gravitropism is consistent with light-stimulated biosynthesis.

figures and graphs with small variations by touch than for others to read them by sight. A wax pencil can be used for one of the bar graphs. A tracing wheel or other tool can be used for the second bar.

#### **VARIATION 2**

The Effect of Light on the Gravitropic Response of Corn Roots

Note to Teachers: In addition to the information found in the Core Experiment, the following material has been provided for Variation 2.

#### **SYNOPSIS**

Students will compare the angle of root growth of corn seedlings after changing their orientation and exposing them to light or keeping them in the dark.

#### ADDITIONAL MATERIALS NEEDED

You will need the following for each group of three students in a class of 24:

- 12 corn seedlings with straight roots about 1 cm long
- 1 light source
- 2 dark cabinets or boxes
- 🕥 1 heat trap
- 1 clear, colorless germination container

#### DIRECTIONS FOR SETTING UP THE EXPERIMENT

Use the setup in Steps 1 to 8 of the Core Experiment to germinate corn seedlings. Do not vary the angle of the seed. Adjust the seeds so that the roots emerge directly down.

#### HYPOTHESIS GENERATION

#### Question

What effect will light from below have on the gravitropic response of corn?

#### Sample Hypothesis

If corn roots are lighted from below, the apparent gravitropic response will be less than if the roots are placed in the dark.

#### Rationale

Roots normally grow in the dark. They may have a negative phototropic response that appears as a stronger gravitational response. Light from below could send the wrong signal.

#### Sample Experimental Procedure

- 1. Divide the roots into matched control and treatment groups.
- 2. Set up the control group.
  - a. Reorient the roots to the perpendicular either by cutting the paper that they have germinated on or by rotating them.
  - b. Make a mark on the paper just at the tip of the root.
  - c. Return the seedlings to their germination chamber.
  - d. Shield the chamber from light by putting it in a dark cabinet or box.
- 3. Set up the treatment group.
  - a. Reorient the roots to a perpendicular position by cutting the paper on which they have germinated and rotating it.



- b. Make a mark on the paper just at the tip of the root.
- c. Place seedlings in a clear, colorless, covered germination chamber.
- d. Shield the chamber from stray light by putting aluminum foil around the top and sides.
- e. Place the chamber immediately over the heat trap.
- f. Place the light immediately under the heat trap and turn it on.
- 4. After 2 hours, draw a dot at the position of each root tip or end and draw a line between the 2 dots marking the initial and end positions.
- 5. Measure and record the angle between a horizontal line drawn through the starting position of the root and the line you drew from that point to the end point. Record values above the horizontal as negative and those below the horizontal as positive.
- 6. Analyze the results of your measurements.

#### DATA ANALYSIS AND INTERPRETATION

#### Sample Data

**Table 5.** Angle of roots reoriented to a horizontal position and incubated with lights placed below or in room light for 2 hours. Measurements are in degrees from the horizontal. Growth below the horizontal position is measured as a positive number. Growth above the horizontal position is measured as a negative number.

Replicate	Root angle (*)	
	Room light incubation	Directional light incubation
1	60	90
2	60	70
3	60	50
4	34	58
5	65	28
6	60	42

Appropriate analysis is the Wilcoxon Two-Sample Test. Here C = 17 and C' = 27. The critical comparison value is 29.

#### TEST QUESTION

Some plants require light to germinate. Roots of these plants do not necessarily exhibit negative phototropic responses. What might be the adaptive value of a light requirement for germination?

#### SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

#### Blind or Visually Impaired

• Students who are visually impaired should have no difficulty with this experiment if they understand the Wilcoxon Two-Sample Test. Some math departments might assist with this.



#### Interpretation

These results do not support the hypothesis. The Wilcoxon statistic is less than the tabled critical comparison value, so one can conclude that differences as great as those observed in these data sets are likely to occur by chance alone.

#### Answer to Test Question

Roots and shoots have only the energy stored in the seed before the shoots reach light and begin to photosynthesize. If a seed can detect light it is likely that the shoot could reach light before running out of energy reserves. If this idea has any merit, one would expect to find a light requirement for germination more common among small seeded species than among large seeded species.



#### VARIATION 3 The Effect of Water on the Gravitropic Response of Corn Roots

*Note to Teachers:* In addition to the information found in the Core Experiment, the following material has been provided for Variation 3.

#### SYNOPSIS

Students will determine if water will alter the gravitropic response of corn roots.

#### ADDITIONAL MATERIALS NEEDED

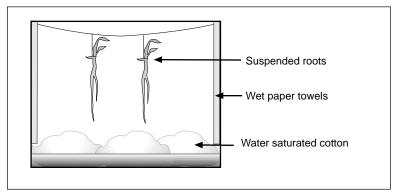
You will need the following for each group of three students in a class of 24:

1 5 x 5 x 2-cm piece of absorbent cotton

- 1 dropping pipette
- 1 transparency film
- 12 corn seedlings with straight roots about 1 cm long

#### DIRECTIONS FOR SETTING UP THE EXPERIMENT

Germinate seeds as in Core Experiment. Do not vary the angle of the seed. Place seed so roots emerge going down.



**Figure 6.** Cross section of a chamber to detect the interaction of hydrotropism and gravity on roots.

#### HYPOTHESIS GENERATION

#### Question

How will corn roots respond to gravity if the water source is not below the roots?

#### Sample Hypothesis

If corn roots are exposed to water that can be reached easily, but that is not located below them, they will not respond as strongly to gravity.

#### Rationale

A tropic response to water could be a mechanism for avoiding drought.

Sample Experimental Procedure

- 1. Divide the roots into 2 matched groups. One group will be the control and the other the treatment group.
- 2. Set up the control by repeating the following steps for each replicate.
  - a. Fasten (glue or staple) a  $5 \times 5 \times 2$ -mm piece of absorbent cotton at the right edge, about 1 cm from the upper edge of a piece of transparency film.

#### TEACHING TIPS

 Research in the 1800's showed that at least some plants grow toward regions of higher moisture. For some roots, hydrotropism can overcome gravitropism (Takahashi, 1994).

 Another possible design would be to line a box with wet paper towels to establish a uniformly humid chamber. In the middle of the chamber, suspend a pair of roots in the arrangement shown in Figure 6. b. Affix a germinated corn seedling to the transparency film with the root horizontal and 1 mm from the left edge of the cotton. See Figure 7.

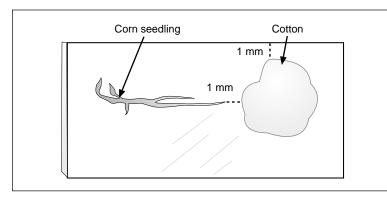


Figure 7. Seedling attachment to transparent film.

- c. Make no additions to the cotton.
- d. Return the plant to the germination chamber.
- 3. Set up the treatment by repeating Steps 2a and b for each replicate.
  - a. Saturate the cotton with distilled water.
  - b. Return the plant to the germination chamber.
- 4. After 4 hours, measure the angle of curvature.
  - a. Mark the position of the root tip.
  - b. Draw a line from that final mark to the initial tip placement.
  - c. Measure and record the angle between a horizontal line through the initial tip placement and the final mark.

#### DATA ANALYSIS AND INTERPRETATION

#### Sample Data

Table 6. Angle of root growth with and without a horizontally supplied water source.

Replicate	Root angle (*)	
	Dry cotton at root tip	Water saturated cotton at root tip
1	60	63
2	60	60
3	60	60
4	65	72
5	55	60
6	60	78

Use the Wilcoxon Two-Sample Test to analyze these data. Here C = 4 and C' = 32. The critical tabled comparison value is 29.

#### TEST QUESTION

Study the directions for performing the Wilcoxon Two-Sample Test for analyzing data like that collected in this experiment. How many samples are necessary to conclude that the differences observed are significant if there is no overlap in these data? Assume that you are content to make an error in your interpretation 5% of the time. The .05 column is all that you have been given. Explain.

#### Interpretation

**TEACHER'S NOTES** 

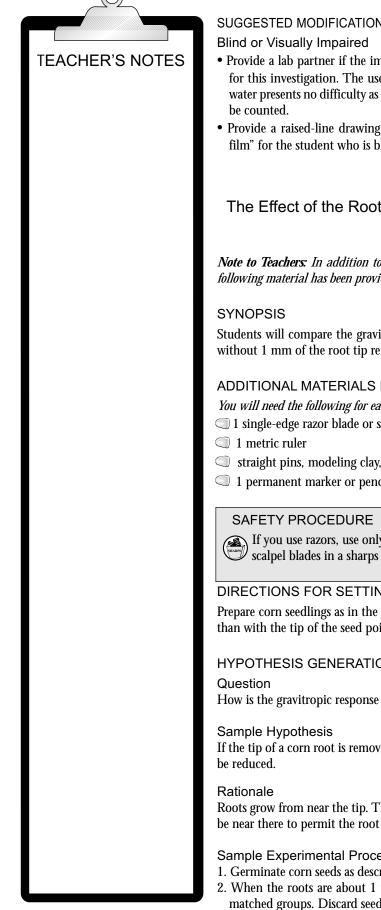
These results do not support the hypothesis. Data sets as different as those found most likely differ by chance alone. One would expect less of a downward growth when water was supplied laterally, but the greatest downward curvature occurred then.

#### Answer to Test Question

Three samples are adequate if there is no overlap. The Wilcoxon statistic calculated will be

C = 3(3) + [3(4)/2] - 6 = 9 and C'= 3(3) - 9 = 0.

The tabled comparison value is 9. Since the calculated value is equal to the tabled value, the differences in these data sets are not due to chance alone.



#### SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

- Provide a lab partner if the impaired student needs assistance handling the roots for this investigation. The use of a dropping pipette to saturate the cotton with water presents no difficulty as long as the number of drops of water do not need to
- Provide a raised-line drawing of Figure 7, "Seedling attachment to transparent film" for the student who is blind.

#### **VARIATION 4** The Effect of the Root Tip on the Gravitropic Response of Corn Roots

Note to Teachers: In addition to the information found in the Core Experiment, the following material has been provided for Variation 4.

Students will compare the gravitropic response of corn seedling tap root with and without 1 mm of the root tip removed.

#### ADDITIONAL MATERIALS NEEDED

You will need the following for each group of two students in a class of 24:

- 1 single-edge razor blade or scalpel
- straight pins, modeling clay, or glue
- 1 permanent marker or pencil

If you use razors, use only those with single edges. Dispose of razors and scalpel blades in a sharps container.

#### DIRECTIONS FOR SETTING UP THE EXPERIMENT

Prepare corn seedlings as in the Core Experiment, but do not orient the seed other than with the tip of the seed pointing downward.

#### HYPOTHESIS GENERATION

How is the gravitropic response of corn affected by the removal of its tip?

If the tip of a corn root is removed, then the gravitropic response of a corn root will

Roots grow from near the tip. The sensors for moisture, obstacles, and gravity must be near there to permit the root to change direction.

#### Sample Experimental Procedure

- 1. Germinate corn seeds as described in Steps 1 through 7 of the Core Experiment.
- 2. When the roots are about 1 to 1.5 cm long, divide the seedlings into 2 closely matched groups. Discard seedlings with crooked roots.



- 3. One group is the control.
  - a. Carefully remove the seedlings from their growing situation.
  - b. Blot each seed dry and fasten it to a fresh piece of paper so the root is perpendicular to its previous position.
  - c. With a pencil or permanent marker, mark the position of each root tip on the paper.
  - d. Completely wet the paper and place it in a germination chamber like the original.
- 4. The second group is the treatment group.
  - a. Carefully remove the seedlings from their growing situation.
  - b. Use a sharp razor blade to cut off the root 1 mm from the tip. See Figure 8.

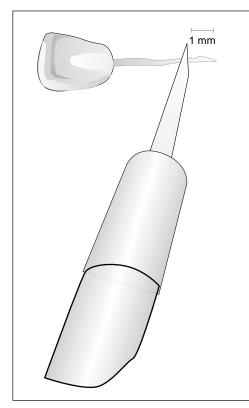


Figure 8. Removal of 1 mm of root tip.

c. Blot each seed dry and fasten it to the paper or transparency film so the root is perpendicular to its previous position. See Figure 9.

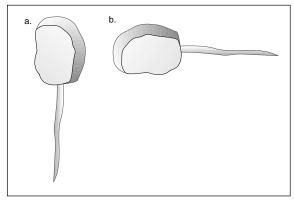
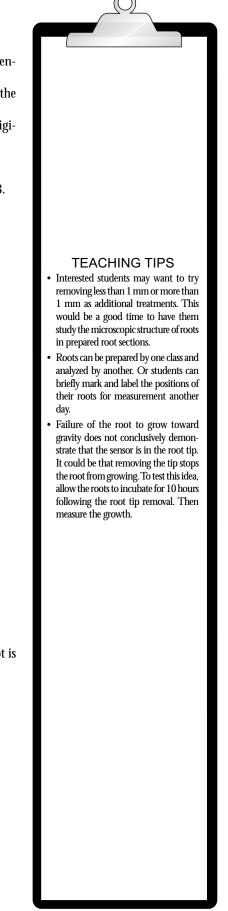


Figure 9a. Original orientation. b. Perpendicular orientation.



#### TEACHER'S NOTES

### d. With a pencil or permanent marker, mark the position of each root end on the paper.

- e. Place the seedlings in the germination chamber with the root horizontal. Completely wet the paper before placing the seedlings in the germination chamber.
- 5. After 4 hours, draw a dot at the position of each root tip or end and draw a line between the two dots marking the initial and end positions.
- 6. Measure and record the angle between a horizontal line and the line you drew. Record values above the horizontal as negative and those below the horizontal as positive.
- 7. Analyze the results of your measurements.

#### DATA ANALYSIS AND INTERPRETATION

#### Sample Data

**Table 7.** Effect of removing 1 mm of root tip on the gravitropic response of cornseedlings over 4 hours.

Replicate	Root angle (degrees from horizontal)	
	Treatment (tip removed)	Control (root intact)
1	-10	30
2	-5	38
3	0	40
4	0	35
5	-5	35
6	0	30
Mean	-3	35

Draw a picture of two seedlings representing the average final root angle. Label the angle with the average degree from horizontal. Appropriate statistical analysis here is the same as in the Core Experiment.

#### TEST QUESTION

Another group of students measured the growth of roots with and without the terminal 1 mm removed. In every other way, they handled the roots as done in this experiment. Graph their results presented in Table 8 and write a conclusion.

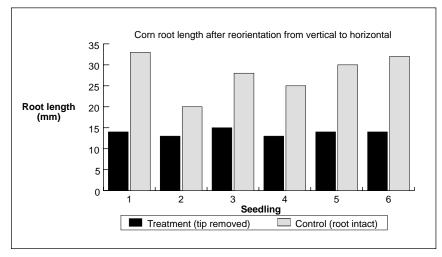
Table 8. Root length 10 hours after roots were reoriented perpendicular to gravity.

Seedling	Root length (mm)	
	Treatment (tip removed) Control (root intact)	
1	14	33
2	13	20
3	15	28
4	13	25
5	14	30
6	14	32

#### Interpretation

The hypothesis is supported by these data. Intact roots had an average positive gravitropic response of 38° more than roots from which 1 mm was removed.

Answer to Test Question See Graph D.



**Graph D.** Corn root lengths were measured 10 hours after they were reoriented from vertical to horizontal. One mm of the tip was removed on half the plants.

#### SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

Blind or Visually Impaired

- At the beginning of the course, discuss the use of razors and scalpels by students who are blind. Those who have used paring knives in the kitchen and those who shave with non-electric razors should have no difficulties.
- Mark positions of roots on the paper with a braille stylus.

#### VARIATION 5

The Effect of Corn Variety on the Response to Gravity

*Note to Teachers:* In addition to the information found in the Core Experiment, the following material has been provided for Variation 5.

#### SYNOPSIS

Students will compare the angle of root growth for different varieties of corn.

#### ADDITIONAL MATERIALS NEEDED

*You will need the following for each group of four students in a class of 24:* Several seeds of a second variety of corn.

#### DIRECTIONS FOR SETTING UP THE EXPERIMENT

Put the seeds of different varieties in water to presoak for 12 hours before affixing them on construction paper.

#### SAFETY PROCEDURES

Handle fungicide-treated seeds with forceps or wear gloves while handling them. TEACHER'S NOTES

#### **TEACHING TIPS**

- Place the seeds of different varieties in the same germination chambers since conditions may vary slightly between chambers.
- If your students are confident that they can orient the corn seedling with the embryo in a horizontal position, they may want to use the design of the Core Experiment.
- Primary roots of corn will show positive gravitropism within 10 to 15 minutes (Lee, Mulkey, & Evans, 1983), so the time schedule for this exercise is flexible. However, measure the root angle made during the first 0.5 to 1.0 cm of growth after reorientation.
- You can save several steps if the roots are straight down. Simply cut the paper on which they have germinated and rotate the piece with the seedling 90°.

#### Interpretation

The mean root angle for Variety A is  $62^{\circ}$ . The mean root angle for Variety B is  $39^{\circ}$ . These data show a strong difference in root growth angle response between Variety A and Variety B. Variety A has a more positive gravitropic response by an average of  $22^{\circ}$ . The gravitropic response of these corn varieties appears to be controlled by genes.

#### HYPOTHESIS GENERATION

#### Question

Would failure to detect a difference in gravitropic response between different corn varieties have provided conclusive evidence that the response is not under genetic control?

#### Hypothesis

If the root angle of corn varieties differs, then the gravitropic response of corn roots is regulated by genes.

#### Rationale

Although corn varieties are not selected for gravitropic responses, that trait is likely linked to a trait for which the corn is bred.

#### Sample Experimental Procedure

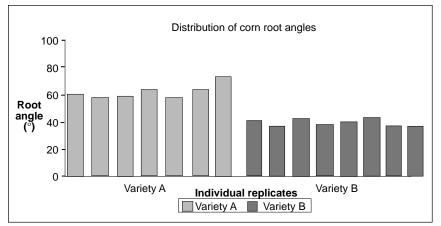
- 1. Draw a line 0.5 cm from the top long edge of the construction paper.
- 2. Affix seedlings of 2 different varieties of corn on construction paper and germinate them as in the Core Experiment. Be certain, however, to set the embryo vertically.
- 3. When the roots are 0.5 to 1.0 cm long, select those with straight, downward pointing roots.
- 4. Work quickly as you handle these seedlings so that the root hairs do not dry. Reorient these roots to the horizontal. They should be parallel to the original line you drew on the paper. Place a mark on the line at the tip of the root.
- 5. After 2 hours incubation in this new position, mark the position of the root tip on the transparency with a permanent marker.
- 6. Draw a line between the tip immediately after reorientation and the tip after further incubation. Measure and record the angle between the horizontal and the line you drew.
- 7. Compare the root growth angle for the 2 varieties.

#### DATA ANALYSIS AND INTERPRETATION

#### Sample Data

Table 9. Effect of gravity on root growth of two varieties of corn.

Replicate	Root angle (°)		
	Variety A	Variety B	
1	60	39	
2	58	37	
3	59	42	
4	61	38	
5	63	40	
6	58	42	
7	63	37	
8	70	37	



Graph E. Effects of gravity on two corn varieties.

#### TEST QUESTION

Would failure to detect a difference in gravitropic response between varieties have provided conclusive evidence that the response is not under genetic control?

#### **VARIATION 6**

The Effect of EDTA on the Gravitropic Response of Corn Roots

*Note to Teachers:* In addition to the information found in the Core Experiment, the following material has been provided for Variation 6.

#### SYNOPSIS

Students will determine if adding a chelator of calcium (EDTA) will influence the gravitropic response of corn roots.

#### ADDITIONAL MATERIALS NEEDED

You will need the following for each group of three students in a class of 24:

- absorbent cotton
- 2 dropping pipettes
- 2 mL of 50 mM EDTA
- transparency film
- 12 corn seedlings with straight roots about 1-cm long

#### DIRECTIONS FOR SETTING UP THE EXPERIMENT

- Germinate seeds as in the Core Experiment.
- To prepare 50 mM EDTA: Place 1.46 g EDTA in 80 mL distilled water. Then add NaOH pellets one at a time to dissolve the EDTA. Dilute to a final volume of 100 mL with distilled water with a pH near 8. Alternatively, use the dihydrate disodium salt of EDTA, which is more soluble. Add 1.86 g to 80 mL distilled water. If necessary, add NaOH pellets to dissolve the EDTA. Adjust the final volume to 100 mL with distilled water.

#### HYPOTHESIS GENERATION

#### Question

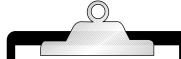
How will the addition of EDTA solution affect the corn root's response to gravity?

#### Answer to Test Question

If we had found no difference in the gravitropic response we could not conclude that the response is not under genetic control. It is possible for two populations to have the same genetic information for some traits. It is also possible that our techniques did not allow us to detect a difference that exists.

#### **TEACHING TIPS**

- Gradients of extracellular calcium are generated across the root tip of corn seedlings and are required for the gravitropic curvature (Bjorkman & Cleland, 1991).
- EDTA (Ethylenediaminetetraacetic Acid) chelates calcium and its effects on the gravitropic response of corn roots can be reversed by supplying calcium (Lee, Mulkey & Evans, 1983).
- Cotton cosmetic pads are excellent material to use here because they make the dimensions easy to control.
- Additional information needed for the test question is given with Variation 2.



Sample Hypothesis

If corn roots are exposed to EDTA, then they will not respond to gravity.

#### Rationale

Calcium has been shown to be associated with corn root gravitropism (Bjorkman & Cleland, 1991).

Sample Experimental Procedure Repeat Steps 1 through 4 of Variation 3 with the following changes: Step 2c. Saturate the cotton with distilled water. Step 3a. Saturate the cotton with 50 mM EDTA.

#### DATA ANALYSIS AND INTERPRETATION

#### Sample Data

Table 10. Root curvature exposed to EDTA or water.

Replicate	Angle of curvature (*)	
	Water (control)	EDTA (treatment)
1	60	0
2	60	0
3	60	0
4	68	0
5	50	0
6	50	0

An appropriate analysis for data collected in this design would be a Wilcoxon Two-Sample Test (Sokal & Rohlf, 1995). These values are so distinctly different with no overlap that no statistical analysis is necessary. No graphical display is needed.

#### TEST QUESTION

A group of students was uncertain whether EDTA would chelate a gravitropically important ion other than  $Ca^{+2}$ . They decided to test whether 30 mM  $CaCl_2$  would reverse the effect of EDTA. If calcium was the only gravitropically important ion chelated by EDTA, what should they find? How could they determine whether the  $Ca^{+2}$  or the  $Cl^{-1}$  was the restorative element?

#### SUGGESTED MODIFICATIONS FOR STUDENTS WHOARE EXCEPTIONAL

Blind or Visually Impaired

- Remind students, especially blind students, to wear rubber gloves when handling seeds treated with fungicide.
- Refer to previous variations for other techniques needed for this variation.

#### Interpretation

EDTA prevented a gravitropic response. The hypothesis is supported by these data.

Answer to Test Question It could be expected that the  $CaCl_2$ would restore the ability of the root to respond to gravity and that the roots would turn downward. They could rule out  $Cl^{-1}$  as the restorative element by testing with MgC1<sub>2</sub> (or MnC1<sub>2</sub>) instead of CaC1<sub>2</sub>. If this compound failed to restore the gravitropic response, then  $Cl^{-1}$  was not the restorative agent.

### **VARIATION 7**

#### The Effect of Light on the Apparent Gravitropic Response of Sunflowers

*Note to Teachers:* In addition to the information found in the Core Experiment, the following material has been provided for Variation 7.

#### SYNOPSIS

Students will determine whether light can overcome the gravitropic response of sunflower shoots.

#### ADDITIONAL MATERIALS NEEDED

You will need the following for each group of four students in a class of 24:

- 10 straight leafy shoots of 10-day-old sunflower seedlings
- 10 100-mL graduated cylinders
- Parafilm<sup>TM</sup>
- light source
- 2 dark cabinets or boxes
- heat trap

#### SAFETY PROCEDURE

Make sure the heat-trap mounts are very stable so there is no danger of spilling water into the light fixture.

#### DIRECTIONS FOR SETTING UP THE EXPERIMENT

- 1. Soak sunflower seeds and germinate in petri dish for 10 days.
- 2. Cut leafy shoots early in the morning. If necessary, remove leaves on some so that all shoots are approximately the same.

#### HYPOTHESIS GENERATION

#### Question

Is the gravitropic response of sunflowers stronger than its phototropic response?

#### Sample Hypothesis

If the gravitropic response of sunflowers is stronger than the phototropic response, then shoots will grow upward whether they are in dark or light.

#### Rationale

Day or night, sunflower shoots grow upward. Gravity is a constant force in the life of a plant, but light is a variable force. Major orientation should be responses to reliable cues. The leaves and flowers track the sun, so it is possible that some of the upward response results from phototropism.

#### Sample Experimental Procedure

- 1. Divide the shoots into 2 matched groups.
- 2. Completely fill the graduated cylinders with water. Use paper towels or cotton to anchor the stems in place with the shoots against the side of the cylinder.
- 3. Gently seal the shoots in the top with Parafilm<sup>™</sup>. See Figure 10.



#### TEACHING TIPS

- Clip-on shop lights are handy light sources in which you can vary the intensity of the light by changing light bulbs. You also can vary the light intensity by changing the distance between the light and the shoots.
- Clear, colorless baking pans make good heat traps when filled with water. They can be supported on two narrow boards over the light source.
- Place the shoot tips against the side of the cylinder so that the half that is not removed will remain in water.
- Shoot growth toward light is controlled by auxin transported from the apical meristem (Laferriere, 1993). With *Coleus blumei*, the effects of gravitropic responses are usually stronger than phototropic responses.
- Tomato seedlings also respond very quickly to gravity and can be easily handled in small pots.
- The results reported here were obtained with shoots 25 cm above a 55-watt incandescent bulb with 3 cm of water in the water trap. If a light meter is available, you may want to quantify the light and test different intensities or qualities of light.

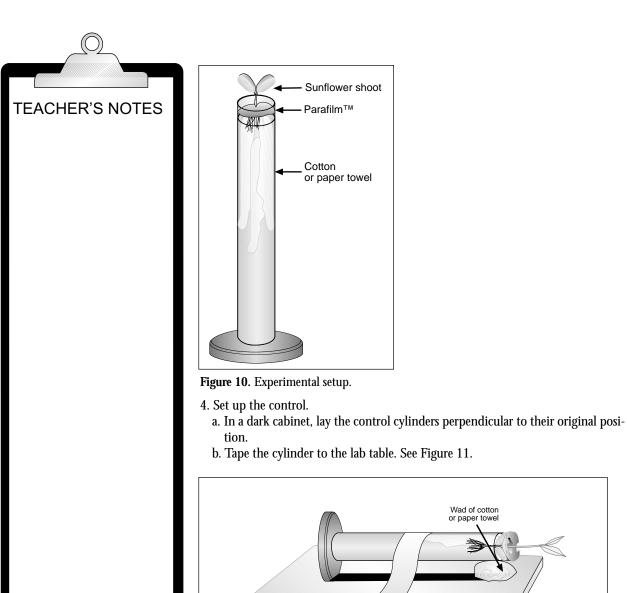


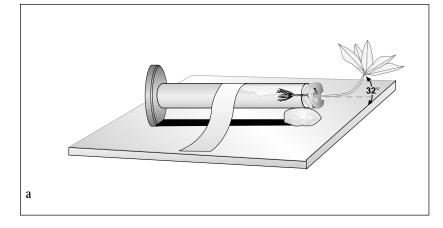
Figure 11. Graduated cylinder taped to the lab table.

- 5. Set up the treatment.
  - a. Lay the treatment cylinder on the lab table in another dark cabinet.
  - b. Immediately below the shoots, place a heat trap.
  - c. Immediately below the heat trap, place the light facing upward.
  - d. Turn on the light.
- 6. After 24 hours, measure the shoot angle from the horizontal. See Figures 12 a & b.

Wad of cotton or paper towel

### DATA ANALYSIS AND INTERPRETATION

#### Sample Data



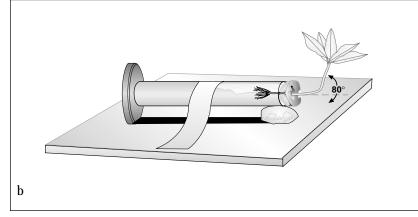


Figure 12 a & b. Stem position after 24 hours in the light (a) and in the dark (b).

 Table 11. Sunflower stem angles after 24 hours exposed to gravity in the dark or lighted from below.

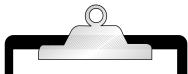
Replicate	Stem angle (*)	
	Light Dark	
1	32	80
2	30	84
3	30	84
4	34	86
5	30	80
6	32	80

An appropriate analysis for this data would be the Wilcoxon Two-Sample Test. Here, however, these data do not overlap and no statistical analysis is necessary. Draw diagrams of curved stems representing the average angle of curvature or chart the distribution of curvature angles.

#### Interpretation

TEACHER'S NOTES

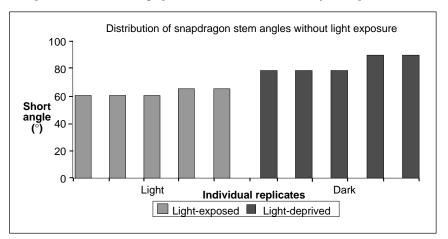
The gravitropic response is stronger than the phototropic response because the angle of curvature is less for shoots lighted from below than for those only exposed to gravity.



Answer to Test Question These stems had rather uniform responses to the treatments. The direction of these responses is the same as those observed; the gravitational influence is stronger than the light response. The snapdragon stems were less responsive to light than were our sunflower stems because they curved less when exposed to the light.

#### TEST QUESTION

Compare the student data graphed below with the results of your experiment.



Graph F. Influence of light on the gravitropic response of snapdragon flowering stems.

SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL Blind or Visually Impaired

• Have the student who is visually impaired note the setups used by sighted students, or team them with a sighted person if braille sketches of the positions of the cylinders and seedlings are not available.

#### **VARIATION 8**

The Effect of Removing the Shoot Tip on the Gravitropic **Response of Sunflower Shoots** 

Note to Teachers: In addition to the information found in the Core Experiment, the following material has been provided for Variation 8.

#### **SYNOPSIS**

Students will compare the gravitropic responses of sunflowers with and without the tip removed.

#### ADDITIONAL MATERIALS NEEDED

You will need the following for each group of two students in a class of 24:

- 1 single-edge razor blade or scalpel
- 10 straight sunflower stems with expanded leaves removed
- modeling clay
- 10 15-mL test tubes
- Parafilm<sup>TM</sup>
- 1 damp paper towel

#### HYPOTHESIS GENERATION

#### Question

How will the gravitropic response of a sunflower be affected if the shoot tip is removed?



#### Sample Hypothesis

If the shoot tip of a sunflower stalk is removed, then the gravitropic response will be less.

#### Rationale

If the sensor for gravitropic response is in the roots, it is likely that the sensor for shoots will be in the tip.

#### Sample Experimental Procedure

- 1. Cut the stems of the 10 sunflower seedlings to the same length and insert them into test tubes filled with water.
- 2. Pack the opening of the test tube with damp paper towels or cotton to anchor the stem and seal the stem in with Parafilm<sup>TM</sup>.
- 3. Divide the sunflower shoots into 2 matched groups of 5. One group will be the treatment group and the other, the control group.
- 4. Place the control plants on their sides with the shoot perpendicular to the floor. Use modeling clay or tape to wedge the test tube in a secure position.
- 5. Trim the shoot apex through the stem as close to the tip as possible.
- 6. Place treatment shoots on their sides with the shoot perpendicular to the floor. Use modeling clay or tape to wedge the pot in a secure position.
- 7. After 2 hours, measure and record the angle of curvature of each shoot. Figure 13 illustrates the angle to measure. Measure the angle formed by a line tangential to the curvature at the stem tip and a line parallel to the initial stem axis.

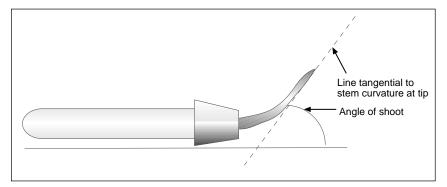


Figure 13. Diagram of technique for measuring the angle of shoot curvature.

#### DATA ANALYSIS AND INTERPRETATION

#### Sample Data

Replicate	Angle of stem curvature	
	Without tip	Intact shoot
1	21	20
2	20	18
3	20	20
4	18	21
5	19	18

 Table 12. Curvature of sunflower shoots with and without tips removed.



#### TEACHING TIPS

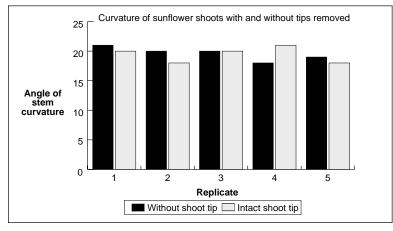
- Shoots with intact leaves will produce the same responses. So much water is transpired by leafy shoots that they are likely to use all the available water and wilt.
- Sunflowers belong to the genus *Helianthus* Small flowered kinds, like Jerusalem artichoke, *Helianthus tuberosus*, provide many useful shoots. You also can use seedlings started from sunflower seed intended for bird feed.
- Without leaves, the shoot will hold over for 24 hours, so the observations could be made the next day.
- The response will be stronger if the leaves are left on the stem, but the loss of water by transpiration will be great. If you want to do this with intact leaves, use only the short time period and replace the straw or narrow glass tubing with a 100-mL graduated cylinder or similarly large water reservoir.
- This response will be observed in a lighted classroom or in the dark. Students may want to perform the experiment in the dark to eliminate the possibility that they are observing a positive phototropism rather than a negative gravitropism.



#### Interpretation

Removing the shoot tip does not alter the gravitropic response of sunflower shoots. The mean control shoot angle was 19.6° and treatment angle height was 19.4°. These data for treatment and control almost completely overlap. The hypothesis is not supported.

Answer to Test Question The stem's main function is to display the leaves to the sun. It is adaptive for the leaf buds to be in a position for the expanding leaf to immediately receive sunlight rather than to have the leaf be the sensor for light or gravity. To orient the leaf buds correctly, the stem needs to grow toward the light before light is perceived. Even a shoot of a deeply buried seed should reach the light by negative gravitropism.



Graph G. Effect of tip removal on the curvature.

#### TEST QUESTION

What is the adaptive advantage for a stem to exhibit a strong negative gravitropic response?

#### SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

Blind or Visually Impaired

• Use straws instead of test tubes with students who are visually impaired.

#### **VARIATION 9**

The Effect of Removing a Dandelion Flower on the Gravitropic Response of the Peduncle

Note to Teachers: In addition to the information found in the Core Experiment, the following material has been provided for Variation 9.

#### **SYNOPSIS**

Students will examine the effect of removing the flower from a dandelion on the gravitropic response of the peduncle.

#### ADDITIONAL MATERIALS NEEDED

You will need the following for each group of two students in a class of 24:

- jumbo drinking straws or 6-mm diameter glass tubing
- modeling clay, cotton or paper towel
- 1 metric ruler
- 12 dandelion plants
- 12 15-mm test tubes
- 1 scalpel or razor blade

#### DIRECTIONS FOR SETTING UP THE EXPERIMENT

- Collect peduncles that are at least 15 cm long which are in the bud stage or stand erect and just beginning to disperse fruits. Keep them fully upright until they are to be used.
- Seal one end of the glass tubes with heat or seal one end of the straws with modeling clay.
- A syringe is useful for filling the glass tubing or straw with water.



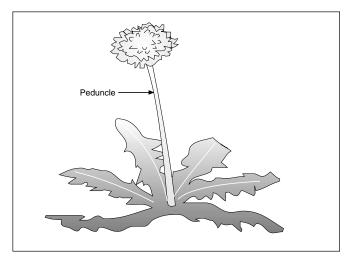


Figure 14. Dandelion plant.

#### HYPOTHESIS GENERATION

#### Question

Will the gravitropic response of the dandelion peduncle be affected if the bud of the flower is removed?

#### Sample Hypothesis

If the flower from a dandelion peduncle in bud stage is removed, the peduncle will not respond to gravity.

#### Rationale

The sensor for root gravitropic response is in the root tip so it seems likely that the sensor for the flower stalk will be at the tip.

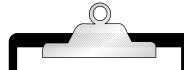
#### Sample Experimental Procedure

- 1. Keep the peduncles upright while you work with them.
- 2. Fill 1 tube with water for each peduncle you have.
- 3. Place the peduncles into the tubes so that at least 10 cm extend above the tube. If the peduncles are dramatically different in length, cut them to about the same size by cutting off the lower portion. Retain the portion closest to the flower.
- 4. Gently anchor the peduncles so they cannot rotate in the tube by wedging in a little modeling clay, cotton, or tissue.
- 5. Divide your prepared peduncles into 2 groups. Leave the flower attached to the control peduncles. Cut the flower head from the treatment peduncles.
- 6. Lay the tubes on a counter so that the peduncles extend beyond the edge and anchor them in place with a small piece of modeling clay or tape.
- 7. After 3 hours, measure the perpendicular distance to the tip of the peduncle or the base of the flower from the plane of the counter on which its base is resting.
- 8. Analyze the growth of the peduncles.



#### TEACHING TIPS

- A graduated cylinder makes a good carrying and storage container for dandelion flowering stalks (peduncles).
- The gravitropic response of dandelion peduncles varies with the stage of development (Clifford & Oxlade, 1991). Peduncles are strongly negatively gravitropic in bud, weakly gravitropic in flower, sometimes diagravitropic when the petals fall and the head closes during fruit maturation, and again strongly negatively gravitropic just before the fruits are dispersed.
- Expect a lag time of 20 to 30 minutes before the peduncle grows upward (Clifford & Oxlade, 1991).
- A 10-cm peduncle may become vertical in 2.5 hours (Clifford & Oxlade, 1991).
- The flower of a dandelion is a cluster of flowers in an inflorescence called a head. The scientific name for dandelion is *Taraxacum officinale*, Weber.
- Some flower stalks are strongly positively gravitropic or thig-motropic when their fruits develop. For example, peanuts (*Arachis*) are buried by the flower stalk, fruit axes of a relative of snapdragons (*Antirrhinum*) creep into fissures, and stems of a relative of rhubarb (*Polygonum*) drop seeds as they creep along the ground (Van der Pijl, 1972)
- Dandelion peduncles will continue to grow for several days if they are kept upright in water. See Figure 14.
- You can demonstrate that the bending of the dandelion peduncle is the result of differential growth by splitting the peduncle longitudinally, separating the top and bottom halves of a bent peduncle. You can measure a difference in the length of these sections. See Figure 14.
- A variation would be to study the height change over time by recording the height at half-hour intervals. There is usually a lag time in the appearance of the curvature.
- Weed seeds can be purchased from: Valley Seed Service
   PO Box 9335
   Fresno, CA 93791
   559.435.2163



#### DATA ANALYSIS AND INTERPRETATION

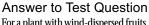
Sample Data

**Table 13.** Effect of removing the flower on the gravitropic response of dandelion peduncles after 3 hours.

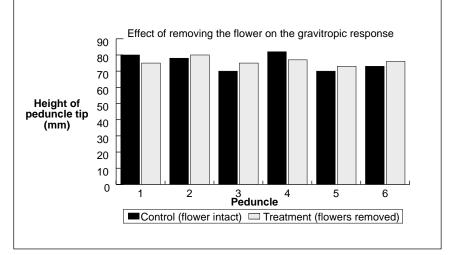
Peduncle	Height of peduncle tip (mm)	
	Control (flowers intact)	Treatment (flowers removed)
1	80	75
2	78	80
3	70	75
4	82	77
5	70	73
6	73	76

#### Interpretation

Removing the flower does not alter the gravitropic response of the peduncle. The mean control peduncle height was 75.5 mm and treatment height was 76 mm. These data for treatment and control overlap almost completely. The hypothesis is not supported.



For a plant with wind-dispersed fruits, erecting the flower stalk well into the air would be advantageous since it could elevate the fruits above surrounding vegetation and place them in a good launching location. Any plant, such as the peanut with its fruit or seed underground, could be expected to be positively gravitropic. A mechanism for orienting to the soil must be present.



Graph H. Flower removal effect on the gravitropic response.

#### TEST QUESTION

Dandelion fruits are wind-dispersed. What is the adaptive advantage for having strong negatively gravitropic flower stalks in this species? Peanut fruits are buried by their flower stalks. Would you expect peanut flower stalks to be negatively gravitropic? Explain.

### SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

Blind or Visually Impaired

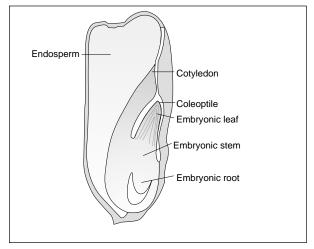
• Remind the student who is visually impaired that the peduncle is the stem. Drawings labeled in braille would be very useful.

# Which Way to Grow

### **Directions for Students**

#### INTRODUCTION

ROOTS CHASE THEMSELVES IN TIGHT CIRCLES! Could this be a news flash from the space station greenhouse? Could it be an everyday occurence there? Here we know this is not the common growth pattern. We might see it at the bottom of a potted plant that has outgrown its container, but the unrestrained growth of roots is normally different. Since the function of stems is to display leaves and the function of roots is to anchor and absorb water and nutrients, the upward orientation of stems and the downward orientation of roots is adaptive. Will plants orient correctly in space? What do you know about how plants perceive their surroundings? Which components of their surroundings do they respond to? Once they perceive the situation, how do you suppose they respond? Do all plants respond the same way? What does adaptive mean? Why are concerns about plant growth different in space than they are on Earth?



# **Figure 1.** The structure of a corn seed. OBJECTIVES

At the end of this lab, you should be able to:

- Explain how gravity is thought to cause the geotropic response in a corn plant.
- Examine the differences in gravitropic responses of corn variety or other plant species.
- Explain the influence of gravity, light, or touch on other plant organs.
- Describe the interaction of two environmental stimuli on plant growth.

#### SAFETY NOTE

Handle fungicide-treated seeds only with forceps or gloved hands.

#### MATERIALS NEEDED

- For each team of two students, you will need the following:
- I package of mounting putty or 12 straight pins
- 12 presoaked corn seeds
- 4 pieces of 10-cm filter paper or 10 x 10-cm chromatography paper
- I plastic food storage box with lid
- I protractor
- I pencil



#### STUDENT LITERATURE SEARCH SUMMARY WITH REFERENCES

Do a literature search on the topic of gravitropism and other tropic responses of plants. Summarize your findings and cite your references. The Internet is a rich resource for this topic because much of the research is government funded in connection with the space program. Your teacher may be able to suggest some references.

#### HYPOTHESIS GENERATION

From the information you have on this topic, develop a hypothesis that could be tested in a controlled experiment that gathers quantitative data. Explain the reasoning behind your hypothesis. Answer the following questions:

- 1. What is the question you are investigating?
- 2. What makes this question an interesting or important topic for investigation?
- 3. What is the variable in your experiment?
- 4. What other variables will you need to control?
- 5. Why is controlling variables important?

#### PLAN OF INVESTIGATION

Make a numbered list of the steps you will use to investigate your topic. Answer the following questions:

- 1. How many samples have you included?
- 2. What will you measure?
- 3. How can you show your results in a graph?

Design an experiment to test your hypothesis. Be sure that you include an experimental control and enough replicates to provide reliable data. Consider how you will analyze and present your results. Write the procedures you will follow.

#### You <u>must</u> have your teacher approve this protocol <u>before</u> you begin this experiment.

#### QUESTIONS AND ANALYSIS

Once you have collected and analyzed your data and graphed your results, answer the following questions:

- 1. Combine your results with those of all your classmates. Construct a graph showing the relationship between the orientation of the embryo and the displacement angle of its root. Describe the relationship.
- 2. Do these data support your hypothesis? Discuss the evidence you used to come to this conclusion.
- 3. How could you create a variety of corn with the greatest gravitropic response you found among your tested kernels?
- 4. What other stimuli might help orient a root?

#### DESIGN OF VARIATIONS OF CORE EXPERIMENT

- What is the effect of increased temperature on the root growth angle of corn seedlings?
- What effect will light from below have on the gravitropic response of corn?
- How will corn roots respond to gravity if the water source is not below the roots?
- How is the gravitropic response of corn affected by the removal of its tip?
- Would failure to detect a difference in gravitropic response between different corn varieties have provided conclusive evidence that the response is not under genetic control?
- How will the addition of EDTA solution affect the corn root's response to gravity?
- Is the gravitropic response of sunflowers stronger than its phototropic response?
- How will the gravitropic response of a sunflower be affected if the shoot tip is removed?
- Will the gravitropic response of the dandelion peduncle be affected if the bud of the flower is removed?

