A Study of Snail Behavior

SYNOPSIS FOR CORE EXPERIMENT
Students will determine if land snails, genus *Helix,* respond to different chemical stimuli in their environment by observing their behavior.

APPROPRIATE BIOLOGY LEVEL
Introductory or advanced

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Directions for Students

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 288. 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:

- Determine how snails respond to chemical stimuli, including chemicals found in their environment.
- Discuss why snails may respond differently to various chemical stimuli in their environment in terms of evolution.

MATERIALS NEEDED

For the teacher preparation, you will need the following for a class of 24:

- 1 10-gallon terrarium or 40 x 22 x 25-cm (16 x 9 x 10-inch) plastic storage box with lid
- 10 g calcium carbonate ($CaCO_3$) chips or 1 cuttlebone
- 500 mL pond or aged spring water
- 1 10-cm jar lid or petri dish cover
- 4 sheets of paper towel or sphagnum moss
- 1 head of lettuce
- 5 spinach leaves
- 3 20 x 20-cm cheesecloth sections
- 10 g glucose
- 1 100-mL graduated cylinder
- 1 balance
- 2 200-mL beakers
- 1 blender

Optional:

- 1 refrigerator
- 1 tea bag
- 5 aged tree leaves, such as oak, maple, or birch
- 17 mL 58% ammonium hydroxide (NH_4OH)
- 5 mL acetic acid (CH_3COOH) or 100 mL vinegar
- 25 mL 40% ethanol (CH_3CH_2OH)
- 10 g sucrose ($C_{12}H_{22}O_{11}$)
- 10 g sugar substitute
- 10 g salt (NaCl)
- 10 g baking soda

LENGTH OF LAB

A suggested time allotment follows:

Day 1 (45 minutes)

- Introduce and observe snails for positive or negative response to environmental chemical stimuli. Brainstorm and develop hypothesis.

Day 2 (45 minutes)

- Design and set up experiment.

Day 3 (45 minutes)

- Analyze and interpret the data.

TEACHER'S NOTES

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

- 1 1-L plastic or glass jar with lid or 1 3 x 10 x 10-cm plastic sandwich container with snap-on lid
- 1 8 x 8-cm piece of 2-cm thick Styrofoam™
- 1 compass with pencil
- 2 25-cm filter paper pieces, paper towels, or coffee filters
- 1 mister/sprayer
- 1 large land snail (*Helix)*
- 1 20 x 30-cm plastic sheet protector, report cover, or glass plate
- 1 25-cm diameter glass pie plate or clear plastic dish
- 1 permanent marker
- 1 metric ruler
- 1 pair of scissors
- 10 mL spring water
- 3 25-mL vials
- 1 forceps
- 1 clock or watch with second hand
- 10 mL 10% spinach extract
- 10 mL 10% glucose solution
- 1 lab journal

SAFETY PROCEDURES

- Use only substances that can be poured safely down the sink to make disposal easier.
- Do not use chemicals that are toxic or dangerous to humans.
- Never use pure caffeine or nicotine. Avoid pesticides and insecticides.
- Check local regulations regarding snails. They may be prohibited in your part of the country, e.g. the transport of snails into Florida and California is prohibited.
- Do not release snails into gardens or greenhouses. Both *Helix aspersa* and *Helix pomatia* are European natives that are serious agricultural pests.
- Do not permit snails to crawl on lab benches.
- Wear safety goggles and aprons when working with chemicals.
- Students should wash their hands before and after working with the snails.
- Keep hands and fingers away from the compass point when punching holes in lids.
- Handle snails with care. Never pull or lift the animal directly from a surface. Rather, gently slide the animal from a surface.
- Always maintain a moist environment. Clean container at least once a week. Do not use distilled water or tapwater directly on snails.

DIRECTIONS FOR SETTING UP THE LAB

Preparing and Maintaining the Snail Habitat

- If you use a plastic storage box instead of a terrarium, punch holes into the lid.
- Line the bottom of the terrarium with the sphagnum moss that came with the snails or sheets of paper towel. Fill the jar lid with pond water or aged spring water and place it in the terrarium. To prevent the snails' shells from becoming brittle, place 10 g of calcium carbonate chips or a cuttlebone in the terrarium.

- Each evening, place a lettuce leaf into the terrarium. Remove any remaining lettuce the following morning.
- The habitat should be placed in a cool location each evening. If possible, place the snails in a refrigerator set to the warmest temperature.
- Replenish the pond water daily.
- Once a week, clean out the habitat by washing the terrarium and jar lid with warm water. Blot the moss to absorb excess moisture and slime, and replace the calcium carbonate.

Preparing the Solutions

Spinach Extract

Puree 2 to 3 spinach leaves in 100 mL of pond or aged water with a blender. Strain the liquid through cheesecloth to remove leaf pieces.

Aged Spring Water

Keep the spring water in an aerated aquarium for a day or more.

10% Glucose, Sucrose, Sugar, Salt, or Baking Soda

Dissolve 10 g of the desired material in a small amount of spring water and dilute to 100 mL with spring water.

Aged Tree Leaves such as Oak, Maple, or Birch (Tannic Acid)

Puree 2 to 3 leaves in 100 mL of water using a blender. Strain the liquid through cheesecloth to remove the leaf pieces.

Tea

Allow 1 tea bag to steep for several minutes in 100 mL boiling water. Cool.

Other substances students may want to test:

10% Ammonium Hydroxide

Add 17 mL of stock (58%) NH_4OH to 83 mL of spring water.

5% Acetic Acid

Add 5 mL acetic acid to 95 mL spring water or use full strength household vinegar.

10% Ethanol

Add 25 mL 40% ethanol to 75 mL spring water.

For lower concentrations of the desired solutions, use the following table.

Table 1. Dilution ratios.

Desired concentration percent	Volume (mL) of 10% solution	Volume (mL) of diluent (spring water)
5	50	50
2	20	80
1	10	90

TEACHER BACKGROUND

Content Information

All organisms must be able to sense the world around them in order to survive. Animals that live in the darkness of caves or the deep sea for their entire lives often have no functional eyes or pigments, but are quite sensitive to chemical stimuli. Bats are sensitive to high frequency sounds that allow them to navigate and find prey in the dark using echo-location. Snakes do not see well, but are able to detect minute amounts of chemicals in the air with specialized "smelling" organs in their mouths. On the other hand, humans have a reduced sensitivity to odors and have come to rely heavily on sight.

Snails are able to sense their surroundings. They need to find food and mates and avoid predators and dangerous situations that not only involve harmful chemicals, but also include potential harm from overheating or dehydration. Land snails, such as *Helix*, sense the environment using two pairs of tentacles. A simple eye is located at the end of each of the longer tentacles. These eyes are able to detect shadows and movement. The shorter pair of tentacles is used for smell and touch. See Figure 1.

PREPARATION TIME REQUIRED

1 hour

- Prepare terrarium for the snails.

30 minutes

- Prepare spinach extract and glucose solution.

TEACHER'S NOTES

When a snail comes in contact with something edible, it will stop and begin eating. The natural food of snails is dependent upon species. If the snail encounters something offensive, it will respond by retracting its tentacles and retreating. Sometimes, a snail even will retract its whole body into its shell. A normal retraction sequence is tentacle invagination, lifting of the anterior part of the foot, folding of that part of the foot along the midline, continued anterior invagination, and finally posterior invagination and withdrawal into the shell. You can use these reactions to test a snail's sensitivity and reaction to various stimuli. See Figure 2.

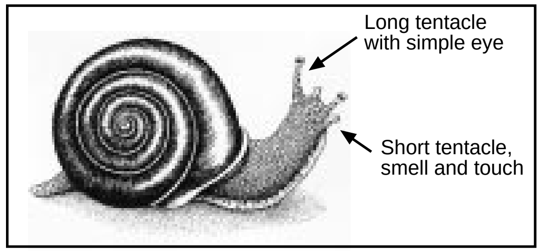


Figure 1. Snail with tentacles' functions identified.

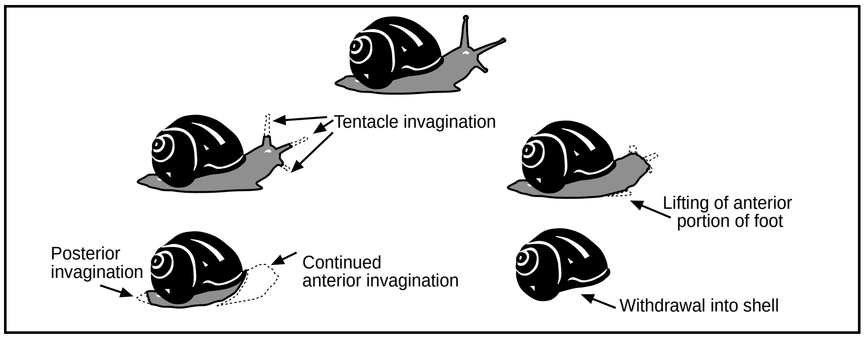


Figure 2. Normal retraction sequence in snails.

Pedagogical Information

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

Correct Concept	Misconception
• Snails respond to chemical stimuli in their environment.	• Snails will not respond to weak chemicals in their environment.
• Snails protect themselves from toxic substances by avoiding them.	• Snails protect themselves from toxic substances by producing slime.

INSTRUCTIONAL PROCEDURES FOR THE CORE EXPERIMENT

Introduction

For an attention-getting device, microwave a bag of popcorn before class. Allow the aroma to penetrate the room so that it greets the students as they enter the room. If you do not have a microwave, open a bottle of cologne or perfume and allow that aroma to penetrate the room.

Have students list the observations they made as they entered class. Invite them to share their answers. Discuss how humans observe their surroundings using the five senses. Ask the following questions as a lead into hypothesis generation:

- What senses do you use most often?
- Which are the most sensitive?
- Why do you need to be able to sense your surroundings?
- When you taste or smell something, what are you actually sensing?
- Do other animals respond to chemicals in their environment?
- How do you know this?

HYPOTHESIS GENERATION

The following discussion and activities are designed to elicit questions that students can transform into hypotheses.

On the board, write a list of up to 10 different animals, including the snail. Include invertebrates as well as some vertebrates. Ask the following questions:

- How does each sense its surroundings?
- Which is the most sensitive to different types of stimuli?
- Which of the listed animals are stimulated by chemicals in their environment?
- What kind of response do they make to chemical stimuli?

At this point, have the students observe snail behavior with a known positive control, such as water, and a known negative control, such as salt solution, so students can define "+" (positive) and "-" (negative) behavior indications. Ask the students to design an experiment to test a snail's response to environmental chemical stimuli. Discuss the importance of testing more than a single animal. Review with your students safe handling of snails and where the snails usually are found so that the hypothesis they suggest has some rational biological basis.

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 287.

Question

Do snails respond to different chemical stimuli found in their environment?

Hypothesis

Snails are able to distinguish between various chemicals in their environment.

Rationale

If an animal is not able to detect common hazards in nature, its chance for survival as an adult decreases.

Procedure: Observation

1. Wash your hands at the beginning of the lab.
2. Obtain a 1-L plastic or glass container or a 3 x 10 x 10-cm plastic sandwich container with a snap-on lid.
3. Place the lid on a piece of Styrofoam™.
4. Punch holes in the lid with the compass point. Be careful not to put hands or fingers where the compass point could puncture them. See Figure 3.

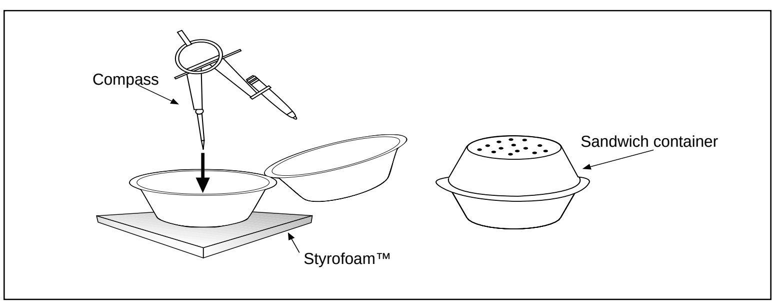


Figure 3. Holes made in sandwich container lid with a compass.

Sample Hypotheses

- Snails will move toward potential food sources that are green and leafy, but will avoid potential foods with animal products.
- Snails will retract into their shells or will move away from offensive chemicals.

TEACHING TIPS

- Tap water may contain substances that will harm snails. Use pond water or aged spring water in the snails' water dish and in the spray bottles. Do not use distilled water.
- The maximum rate of locomotion for the related snail (*Limax maximus*) occurs at about 15˚C and decreases at higher temperatures (Jones, 1975). Moving the snails to a warmer location approximately one–half hour before class will stimulate the snails to become more active.
- Before returning the snails to the terrarium at the end of the lab, rinse them in spring water to remove any chemical residues and blot them dry. Place the snails on the calcium carbonate.
- If the experiment takes longer than one class period, have the students label their snail before returning it to the terrarium so that the same snail is used throughout the experiment. Have students dry the snail's shell and label it with a permanent marker, paint pen, or nail polish.
- Students may become very attached to their snails. Remind them that even though they are studying aspects of behavior and senses, there is no evidence that a retreating strategy, or any other behavior, is planned or that the snails have emotions.
- Deciding how far away to set baits may be difficult. You might want to share the following information about *Helix* or other snails summarized from Hyman (1967) with your students or when you guide them in their selection of materials and concentrations. *Note:* Some of these observations require removing tentacles. Although *Pulmonate* snails have a great ability to regenerate almost any part with the exception of the central nervous system, this procedure is not recommended at the high school level. See Table 2, p. 284.

TEACHING TIPS

SENSE	OBSERVATIONS
Vision	• Dim perception at 1cm. • Distinct vision at 1.5-2 mm. • Movement of objects does not change perception. • Perceive shadows passing over shell and withdraw.
Touch	• Entire surface is sensitive. • Foot is thigmotactic.

Table 2. Observations of snail senses.

A decrease in response to mechanical shock or vibration, light, and moving shadow has been observed in snails stimulated 20 times (Kerkut & Walker, 1975). Snails recovered in about five minutes and showed no signs of being trained. Students need to allow ample time for snails to recover from stimulation before retesting.

- Snails do not learn well, but students should be able to follow some work on this topic summarized by Hyman (1967).
- If some students are reluctant to handle the snails, they can wear plastic gloves.
- *Helix* hibernates from fall to March or April (Hyman, 1967). It may be difficult to work with them during this period as they estivate when it is dry.
- While your students are becoming acquainted with their snails, have them monitor the amount of lettuce eaten per snail. This background information may be useful for Variation 6.
- According to Behringer (1989), a 40 x 22 x 25-cm (16 x 9 x 10-inch) container is adequate for 25 to 30 snails.
- When ordering the snails, specify snails with shells at least 2.5 cm at the largest dimension.
- Provide a calcium source, such as Tums®, calcium carbonate, or cuttlebones.

5. Place a folded piece of 25-cm filter paper, paper towel, or coffee filter on the bottom of the container.
6. Mist with spring water to moisten.
7. Obtain snail and place in the container.
8. Observe the snail within the container.
9. Remove the snail from the container and place on a plastic sheet protector, report cover, or glass plate.
10. Observe the snail once more, especially the behavior of the 2 sets of tentacles.
11. Record your observations.

Procedure: Experimentation

1. Mark the bottom of a 25-cm glass pie plate or clear plastic dish into 2 equal sections with a permanent marker. See Figure 4.
2. Fold the 25-cm piece of filter paper in half.
3. Inscribe a circle with a 10-cm radius with the compass as shown in Figure 5.
4. Cut the outer edge of the arc from the filter paper.
5. Cut the remaining piece of filter paper into two equal halves. See Figure 6.
6. Mark each piece with a pencil as follows:
   a. Water
   b. Dry
7. Pour 10 mL of spring water into a 25-mL vial.
8. Dip one piece of filter paper into spring water with forceps. Leave the other piece dry.
9. Place the filter paper pieces in the dish, equidistant from one another. The pieces of filter paper should not touch. See Figure 7.
10. Place a snail in the center of the dish as shown in Figure 8. Allow it to acclimate to this environment for two minutes.
11. After two minutes, record the number of times that the snail touches each piece of filter paper at 15-second intervals.
12. Continue recording at 15-second intervals for five minutes or 20 counts.
13. Repeat Steps 8 to 12 with spring water. Replace the dry filter paper with spinach extract and then glucose solution.
14. Display your data in a graph and compare them to the results of your classmates.

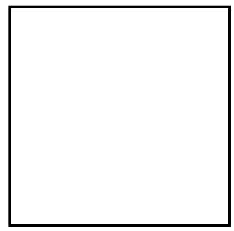


Figure 4. Glass dish divided into two equal halves.

Figure 5. Inscription of circle.

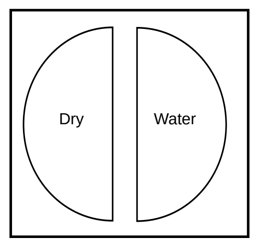


Figure 6. Two filter paper halves.

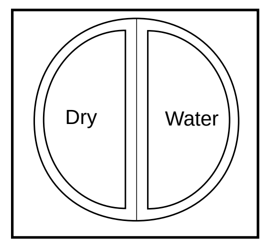


Figure 7. Placement of filter paper halves in dish.

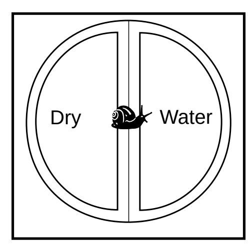


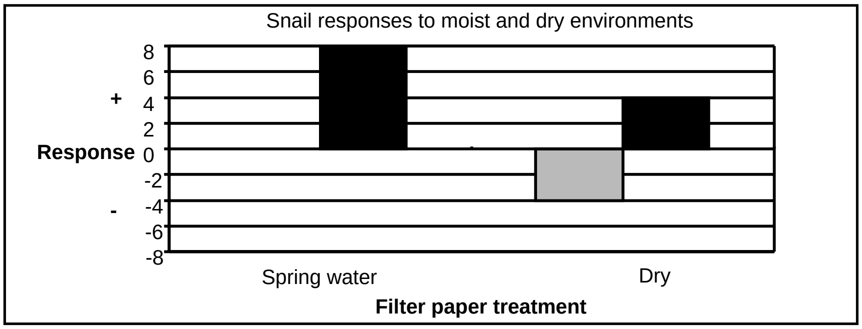
Figure 8. Snail placement in dish.

SAMPLE DATA ANALYSIS AND INTERPRETATION

Sample Data 1

Table 3. Responses of snails to moist and dry environments.

Filter paper treatment n=8	Number of snails with negative response	Number of snails with positive response	Comments
Spring water	0	8	Enters
Dry	4	4	Enters or wanders



Graph A. Snail responses to moist and dry environments.

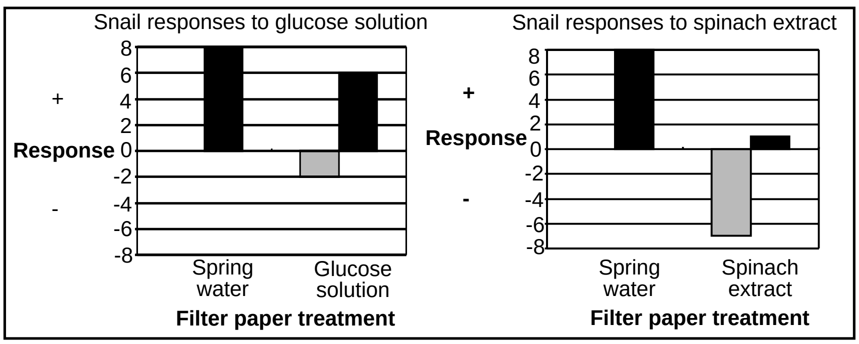
Interpretation

These data support the hypothesis. The snails responded to dry and moist environments by moving into or avoiding the dry environment. The snails can distinguish among dry and moist environments. From an evolutionary point, water is a basic requirement of life.

Sample Data 2

Table 4. Response of snails to environmental chemicals.

Filter paper treatment n=8	Number of snails with negative response	Number of snails with positive response	Comments
Spring water	0	8	Enters
Spinach extract	7	1	Enters or wanders
Glucose solution	2	6	Enters or wanders



Graphs B1 and B2. Snail responses to environmental chemicals.

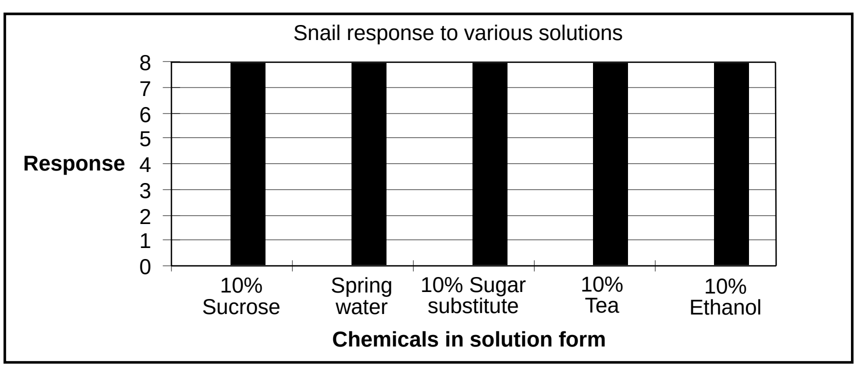
Interpretation

These data do not support the hypothesis. These snails apparently could not distinguish between these chemicals or prefer them equally. All snails moved toward the chemicals and onto the paper with the test solution.

Sample Data 3

Table 5. Response of snails to various chemicals

Filter paper treatment n=8	Number of snails with negative response	Number of snails with positive response	Comments
10% Sucrose	0	8	Enters
Spring water	0	8	Enters
10% Sugar substitute	0	8	Enters
10% Tea	0	8	Enters
10% Ethanol	0	8	Enters



Graph C. Snail responses to single concentrations of various chemicals.

Interpretation

Snails have a positive response to moist environments, as they need moisture to keep sensitive membranes from dehydrating.

Answer to Test Question

A positive response involves movement in the direction of the chemical stimulus; often the foot is stretched as the snail advances. A negative response may be mild, with the snail turning away from the chemical, or strong with the snail lifting its head from the surface before withdrawing into the shell or retreating. We interpreted a lack of movement as a moderately negative response. Snails have a positive response to the control water.

TEST QUESTION

Describe positive and negative response behaviors a snail might exhibit to a particular chemical.

STUDENT DESIGN OF THE NEXT EXPERIMENT

After the students have collected and analyzed the 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 about questions that occurred to them as they conducted the Core Experiment. Ask them what quantifiable experiments could be done based on observations they have made.

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

- What did you observe in your experiment?
- As you were conducting your experiment, what questions came to mind?
- What other factors—chemical, physical, environmental—might influence a snail's behavior? Consider the following:
  -predators
  -food choice and its location
  -effect of light with respect to diurnal versus nocturnal.

SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

These are possible ways to modify this specific activity for students with special needs, who 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 page 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

- Respect the visually impaired or blind student who may or may not be willing to share his/her experience with using taste as a sensory device for the discussion of how humans observe their surroundings. When a sense is absent, it is often said other senses are heightened or more carefully observed.
- Label laboratory shelves of chemicals and labware in braille. The bottles of chemicals and their solutions should be labeled in braille and large print. A strip of sand paper should be attached to hazardous chemicals as a warning. Students who are visually impaired can make minimal observations by touch in an experiment using snails as the primary animal. It will not be possible for them to observe the action of the two sets of tentacles.
- Be aware that students who are visually impaired can make minimal observations by touch in an experiment using snails as the primary animal. It will not be possible for them to observe the action of the two sets of tentacles.
- Provide a copy of the printed laboratory instructions in braille for students who use this reading mode.
- Note that students who are visually impaired should have no difficulty in handling the 25-mm filter paper, placing the halves in a pie plate, and changing solutions as long as materials are accessible.
- Provide accessible, well-labeled solutions so that the student who is blind will have no difficulty in changing the different filter paper sections soaked in various chemicals.
- Assign the laboratory report to be typed by the student who is visually impaired. It should contain the same items that are required of other class members. Bar graphs should be made on braille graph paper. (See "Allelopathy," Variation 1, p. 33).

Deaf or Hard-of-Hearing

- Same considerations as for "Allelopathy," p.29.

Gifted

- One variation that relates well to the garden environment where *Helix* is found is the snail's response to variations in fertilizer concentration.
- Another variation that could bring in skills from physics would be to test the response of *Helix* to additional weight. *Helix* can carry 9 times its weight vertically, but 50 times its weight horizontally (Taylor, 1914 in Jones, 1975).

Manually Impaired

- Be aware that depending on the severity of the impairment, some students will not be able to handle snails or chemical solutions. They can observe the location and behavior of the snails, and report on the position of the tentacles and time actions of snails.
- Assign their laboratory reports to be tape recorded, unless they have a computer with a voice-activated printer. Graphs need to be understood and recalled, rather than drawn.
- Emphasize that the snails should be handled with care. The snails never should be pulled or lifted directly from a dry or wet surface, but be slid along the surface until they move readily.
- Use a light probe to locate and follow the snail in the dish.

Mobility impaired

- Same considerations as for "Allelopathy," p. 30.

ADDITIONAL TEST QUESTIONS

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

1. List the chemicals that elicited a negative response in the snail.
2. Ants are able to travel great distances from their nest to obtain food. They have an unerring ability to always return to their nest if their trail has not been disturbed. This indicates the ant's ability to:
   A. detect gravity.
   B. use the sun as a guide.
   C. follow chemical stimuli.
   D. use magnetic fields as a guide.

REFERENCES AND SUGGESTED READINGS

Behringer, M.P. (1989). *Techniques and Materials in Biology: Care and Use of Living Animals, Plants and Microorganisms* (2nd ed.). Malabar, FL: Robert E. Krieger Publishing Company.

Cain, A.J. (1983). Ecology and ecogenetics of terrestrial molluscan populations. In W. D. Russell-Hunter (Ed.), *The Mollusca: Ecology*, 6. New York, NY: Academic Press, Inc., 597-647.

Child, S.Z., Hartman, C.L. & Shery, L. (1990). A test for increased lethality in land snails exposed to 60Hz magnetic fields. *Neuroscience Letters, 14*(2), 169-170.

Davies, S. (1993). The joy of snails. *BBC Wildlife, 11*(9), 27.

Desbuquois, C. & Daguzan, J. (1995). The influence of ingestive conditioning on food choice. *Journal of Molluscan Studies, 61*(3), 351.

Dussart, G. (1989). Slugs and snails and scientists' tales. *New Scientist*, *123*(1674), 37-41.

Hyman, L. H. (1967). *The Invertebrates: Mollusca I*, 6. New York: McGraw-Hill Book Company.

Jones, C. G. & Shachak, M. (1994). Desert snails' daily grind. *Natural History, 103*(8), 56-61.

Jones, H.D. (1975). Locomotion. In V. Fretter & J. Peake (Eds.), *Pulmonates: Functional Anatomy and Physiology,*1. New York, NY: Academic Press, Inc., 1-32.

Kela, S.L. & Bowen, I.D. (1995). Control of snails and snail-borne diseases. *Pesticide Outlook, 6*(1), 22-27.

Kerkut, G.A. & Walker, R.J. (1975). Nervous system, eye and statocyst. In V. Fretter & J. Peake (Eds.), *Pulmonates: Functional Anatomy and Physiology*, 1. New York, NY: Academic Press, Inc., 165-244.

Answers to Additional Test Questions

1. Answers will vary depending on class observations. A possible list may include: very strong salt, alcohol, ammonia, and/or vinegar.
2. C

Answers to Questions and Analysis on Student Page

1. These graphs should resemble Graphs B1, B2, and C.
2. The answer to this question will depend on the data. If the snails consistently avoid some material and move toward others, accept the hypothesis. If the snails go to every choice in approximately equal numbers, reject the hypothesis.
3. The previous recent experience of the snails might influence their preference; they may prefer a familiar substance. Hunger may influence its selection; a snail that has recently eaten may be more selective than one that is well fed. The choices offered might influence a snail's choice. If it has to choose between two things it does not like, it still may choose something it does not like. It would be useful to know whether any "no choice" options are provided for the snail; that is, are there places it can go simply to escape being tested? Sampling error also could produce the differences seen. It would be useful to know the number of animals that were tested in each experiment.
4. Knowing the sample size certainly could influence confidence in the conclusion. If 4 of 4 animals made one choice, that would be 100% of the animals tested, but the sample size is so small that chance could account for the results. If 24 of 30 snails make one choice, chance would be accepted less likely as an explanation. It could be concluded that these snails have a preference.
5. To test whether odor influences the movement of a snail toward the material, disguise some frequently selected material and treat it with an aromatic material. To focus the source of the odor, put the snail in the middle of a cylinder and put a choice at each end of the cylinder. Test several snails that exhibit an attraction toward the untreated material.

TEACHER'S NOTES

Machin, J. (1975). Water relationships. In V. Fretter & J. Peake (Eds.), *Pulmonates: Functional Anatomy and Physiology*, 1. New York, NY: Academic Press, Inc., 105-163.

Mølgaard, P. (1986). Population genetics and geographical distribution of caffeic acid esters in leaves of *Plantago major* in Denmark. *Journal of Ecology*, *74*,1127-1137.

Runham, N.W. (1975). Alimentary canal. In V. Fretter & J. Peake (Eds.), *Pulmonates: Functional Anatomy and Physiology*, 1. New York, NY: Academic Press, Inc., 53-104.

Stouffer, M. (1988). A multitude of molluscs. In Marty Stouffer Productions, Ltd. (Producer), [Comar, 800.615.4180, distributor), *Animal Oddities*. Aspen, CO: Marty Stouffer Productions, Ltd.

Thompson, R. & Cheney, S. (1996). *Raising Snails*. USDA Special Reference Brief NAL-SRB, shipping list number 96-0350-P. Beltsville, MD: United States Department of Agriculture.

POSSIBLE SOURCES OF MENTORS

United States Department of Agriculture, Beltsville, MD

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

VARIATION 1

The Effect of Spinach Extract and Glucose Dilutions on Snail Response

SYNOPSIS

Students will determine if changing the concentration of the spinach extract solutions will affect the snail's response to them.

ADDITIONAL MATERIAL NEEDED

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

- 1 calibrated dropping pipette
- 1 5-cm vial
- 0.5 mL spinach extract
- 0.5 mL 10% glucose solution

HYPOTHESIS GENERATION

Question

How will changing the solution's concentration affect the snail's response?

Sample Hypothesis

Changing the concentration of the chemical solutions will not change the snail's responses.

Rationale

You expect that at some low concentration most chemicals will not be harmful and the ability to detect them should be low. Above the first level of detection, you might expect no change in the intensity of the response. If a snail is starving, it may consume some materials in excess that can be harmful. So, you may find exceptions. You may reject or accept a hypothesis in this form if you choose to do a statistical analysis. See Variation 4 for directions on statistical analysis.

Sample Experimental Procedure

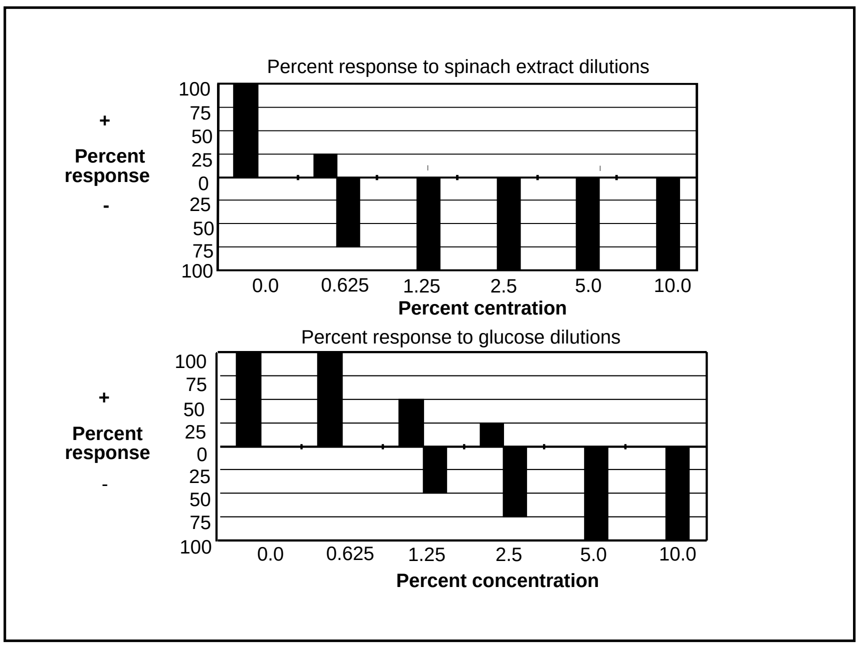
Repeat the Core Experiment procedure, but alter the concentrations of the solutions.

SAMPLE DATA ANALYSIS AND INTERPRETATION

Sample Data

Table 6. Responses of snails to various concentrations of chemicals used in the Core Experiment are shown above. Reported values are number of snails showing a positive response in the numerator and number of snails showing a negative response in the denominator.

	Percent response at various concentrations					
Chemical	0	0.625	1.25	2.5	5.0	10.0
Spinach extract dilutions	4/0	1/3	0/4	0/4	0/4	0/4
Glucose dilutions	4/0	4/0	2/2	1/3	0/4	0/4



Graphs D1 and D2. Snail responses to varying concentrations of spinach extract and glucose solution.

TEACHING TIPS

- Use lower concentrations of the original tested solutions if the response was negative; use higher concentrations if the original response was positive.
- Provide solutions or instructions and pipettes for making solutions of varied concentrations of spinach extract-glucose solutions, such as 1.5, 2.5, 5.0, and 10%. Make the differences 50% to an order of magnitude. You may extend the range of concentrations tested by providing a more concentrated solution as well.
- This is a good place to explain how to make dilutions and have students prepare their own solutions. To make the dilutions used to generate the sample data, use a calibrated dropping pipette to add 0.5 mL of a solution into a 5-cm vial. Then, add 0.5 mL of spring water. The next dilution is made by transferring 0.5 mL of that dilution into another small vial and adding 0.5 mL of spring water. Each solution is half the concentration of the preceding solution. See Figure 9.

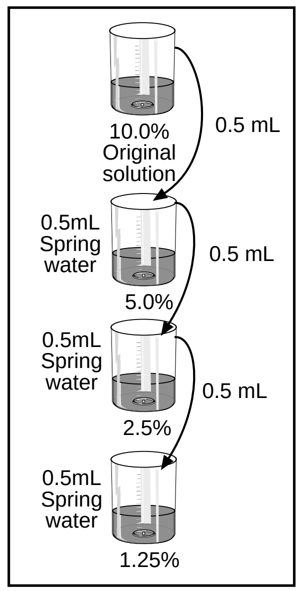


Figure 9. Serial dilution.

Interpretation

Reject the hypothesis if the snails show negative responses to some concentrations and positive responses to other concentrations. Accept the hypothesis if the snails show the same response to all concentrations of the chemicals.

Answer to Test Question

Snails are able to detect differences in concentrations of the chemicals tested. They responded negatively to concentrations of ammonium hydroxide greater than 0.625%, but did not exhibit a negative response to lower concentrations. They also responded negatively to concentrations of alcohol that were equal to or higher than 1.25%.

TEACHING TIPS

- Some species of *Helix* eliminate nitrogenous waste, such as ammonia (Machin, 1975). You might expect that they will show some tolerance for low concentrations of ammonia.
- If the test chemical has a strong odor, begin tests with the lower concentrations and keep the concentrated solutions covered on a distant lab bench. Consider treating everything as though it has a strong odor, since you have not determined whether a snail's olfactory acuity is similar to ours.
- Analyze results as percent responding in a particular way to make data clear when sample sizes are not the same.
- You can cut cardboard sections at the desired inclination angle and substitute these as templates for the protractor with plumb line. A string and weight may be attached to the protractor to act as a plumb line. See Figure 10.
- Snails have two small, liquid-filled statocysts approximately 200 µm in diameter. When the animal is upside down, the statoliths stimulate hair cells in the equilibrium organs called statocysts. These organs are not stimulated in other positions (Kerkut & Walker, 1975).
- Snails climb upward over the range of 15˚ to 75˚. Within this range, they can detect a difference of 2˚. If the climbing plane is rotated, so that the snail becomes briefly horizontal, the time needed for reorientation depends on the angle of inclination. At 90˚, 2.88 seconds are needed; but at 5˚, 12.15 seconds are needed (Hyman, 1967).
- You can substitute plastic for the glass plate, but snails adhere better to the glass.
- Modeling clay can be used to anchor the plate glass at the desired angle.

TEST QUESTION

Study Table 7 below and answer the following questions. Are snails able to detect different concentrations of chemicals? What evidence supports your answer?

Table 7. Responses of snails to various concentrations of chemicals. As in Table 6, reported values are number of snails showing a positive response in the numerator and number of snails showing a negative response in the denominator.

Chemical	Percent response at various concentrations					
	0	0.625	1.25	2.5	5.0	10.0
Ammonium hydroxide	4/4	1/3	0/4	0/4	0/4	0/4
Ethanol	4/0	4/0	2/2	1/3	0/4	0/4

SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

Deaf or Hard-of-Hearing

- Prepare solutions as done in "Allelopathy," Variation 1, p. 33 if not prepared by the instructor.

VARIATION 2

The Effect of Gravity on Snail Behavior

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 determine whether snails are positively or negatively geotactic.

ADDITIONAL MATERIALS NEEDED

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

- 1 10 x 10-cm glass plate
- 1 protractor

HYPOTHESIS GENERATION

Question

What type of geotactic response do snails exhibit?

Sample Hypothesis

Snails will exhibit a negative geotactic response.

Rationale

Helix eggs are laid below ground, and the hatchlings need to reach food above ground. Snails are primarily active at night when light is not available, but gravity is. Movement in the direction opposite the force of gravity is a negative geotactic behavior.

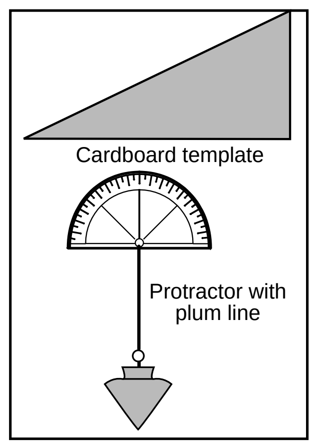


Figure 10. Mechanisms to determine inclination angle.

Sample Experimental Procedure

1. Set a snail at the center of a glass plate in an orientation parallel to the ground. Mist the glass lightly with spring water.
2. Slide the glass to the edge of the lab bench. Position the protractor or inclination angle template against the edge of the plate at the snail's posterior.
3. Gently tilt the plate upward to the desired angle. Measure the angle of inclination as a deviation from the true horizontal as shown in Figures 11a and 11b.

a. b.

Figure 11a. Setup with cardboard template for angle of inclination. Figure 11b. Setup with protractor and plumb line for angle of inclination.

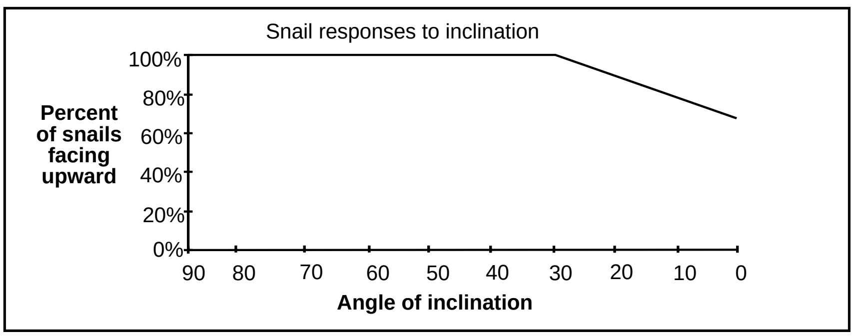
4. As soon as the snail has moved clearly to reorient its head, record whether the new orientation is head up or down. Then, return the glass plate to a horizontal orientation.

SAMPLE DATA ANALYSIS AND INTERPRETATION

Sample Data

Table 8. Responses of snails to inclination.

Angle (˚) of inclination	Number of snails facing upward	Number of snails facing downward
90	8	0
45	8	0
22	8	0
11	8	0
5	6	2



Graph E. Snail responses to inclination.

TEST QUESTION

Why do you usually find the snails you placed on the bottom of the terrarium clinging to the cover in the morning?

SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

Blind or Visually Impaired

- Have visually impaired students participate as a part of a group in this experiment. Some may have or can acquire a braille protractor that can have a plum line attached. If so, they should be able to determine the angle of inclination.
- Tape the edges of the glass plate if it substituted for a plastic plate.
- Collect data in braille for use in the lab report.
- Provide commercial models of snails, if they are available. Lab partners, lab assistants, and friends are often willing to make models if they are aware of the need. They also may produce raised-line drawings and braille paintings.

Interpretation

Reject the hypothesis if the snails climb upward more often than downward or do not climb in any particular direction. Accept the hypothesis if the snails climb downward more often than upward.

Answer to Test Question

Snails that are active during the night can sense up and down. They move upward most often during experimentation. The underside of a leaf is usually more tender than the upper surface where a thicker cuticle protects against water loss. The tender underside would be easier for the snail to eat than the upper surface. If a snail is on the under surface of a leaf, it is probably well hidden from predatory birds. Off the forest floor, it also may be somewhat protected from predatory mice.

TEACHING TIPS

- *Helix aspersa* is negatively phototactic, moving away from a light source (Kerkut and Walker, 1975). The light intensities demonstrated in this reaction are not specified. The direction that light falls on the photoreceptors—sense organs that respond to light—at the ends of the dorsal tentacles, may be important in determining the response. There are no other photoreceptors on *Helix*.
- Most snails and slugs are crepuscular, preferring the low light of dusk, or nocturnal (Cain, 1983). Sometimes, *Helix pomatia* gather in a lighted area (Hyman, 1967). The greatest time of activity is from midnight to 3 a.m. Some cave-dwelling snails show similar diurnal rhythms. You may find different activity levels in an 8 a.m. class and a 1 p.m. class.
- You can create a heat trap by putting a clear, colorless cake pan of cold water between the light source and the snail tubes.
- You can expand this variation by using layers of screening or gray report covers to create a series of increasingly dark regions along a tube. Use a camera light meter to quantify the amount of light under each treatment. You might want to randomize the arrangement of the treatments or use a pair-wise arrangement, such as the design used in the next variation.

VARIATION 3
The Effect of Light on Snail Behavior

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 snails show positive or negative phototactic behavior.

ADDITIONAL MATERIALS NEEDED
You will need the following for each group of two to four students in a class of 24:
- 1 lamp, light bank, or bright window not in direct sunlight
- 1 15 x 15-cm piece of clear, colorless transparency film or report cover
- 1 15 x 7.5-cm piece of black construction paper or opaque black plastic
- 2 rubber bands

HYPOTHESIS GENERATION
Question
What form of phototactic response do snails exhibit?

Sample Hypothesis
Snails will exhibit a negative phototactic response.

Rationale
Snails are active mostly at night. During daylight, it is usually warmer and drier. The loss of water required to remain cool in the light and warmth of day could be great. Movement away from light is a negative phototactic behavior.

Sample Experimental Procedure
1. Use a permanent marker and ruler to draw a line across the middle of the transparency film. Lay the marked side down to avoid any problems with potentially toxic ink.
2. Place your snail on the line orienting the body along the line.
3. Lightly mist the transparency film with spring water. See Figure 12, Step 1.
4. Roll the transparency film into a tube about 4 to 5 cm in diameter and fasten it with a rubber band. See Figure 12, Step 2.
5. Quickly roll black construction paper or opaque plastic over one half of the tube and fasten it with a rubber band. See Figure 12, Step 3.

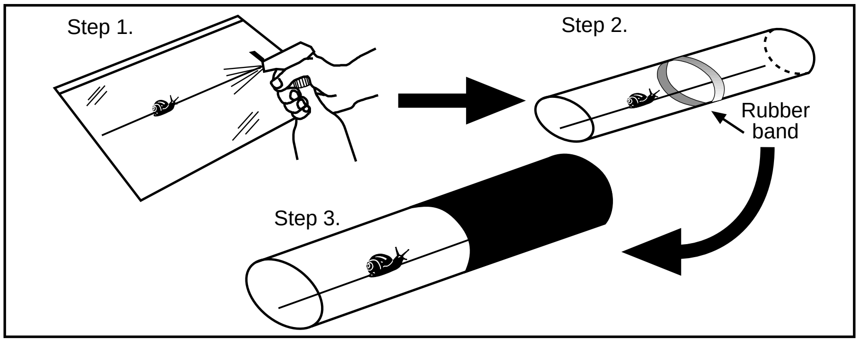


Figure 12. Procedure steps.

6. Lay the tube under the lights. Do not put it close to an incandescent bulb because the temperature can become dangerously high for the snail.
7. Observe the snail's behavior at 1-minute intervals for 10 minutes.
8. Record the direction of the snail's response as "+" if it moves toward the bright half of the tube or as "-" if it moves toward the dark half of the tube.
9. At the end of 10 minutes, disassemble the setup and return the snail to its original container.
10. Use a damp paper towel to clean slime from the snail side of the transparency film and repeat your observations with another snail.

SAMPLE DATA ANALYSIS AND INTERPRETATION

Sample Data

Table 9. The phototactic behavior of snails.

	Positive response	Negative response
Number of snails	0	10

Data as overwhelmingly clear as these require no further analysis. If you want to create a graph, use a bar graph with light treatment on the x-axis and number of snails or percent of responding snails on the y-axis.

TEST QUESTION

What time of day do you expect snails to be active? How do the results of this experiment support your answer?

SUGGESTED MODIFICATION FOR STUDENTS WHO ARE EXCEPTIONAL

Blind or Visually Impaired

- Provide a counter or place for a table underneath a window, if available. It is a better place for the student who is blind to place his/her tubes. The student who is visually impaired then can use a light sensor to follow the location of the snail in the tube.

Interpretation

Reject the hypothesis if the snail consistently moves toward the light or shows no definite directional preference. Accept the hypothesis if the snail consistently moves away from the light source.

Answer to Test Question

The snails all moved away from the light, so they probably are active at night. A range of light intensities was not tested. They may move from very bright light to less light such as from the top of a leaf in direct sunlight to the bottom of a leaf, and actually be active during the day. An extension of this experiment would be to test snail responses to a range of light intensities.

VARIATION 4

The Effect of the Color of Light on Snail Movement

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

SYNOPSIS

Students will determine if snails prefer light of a particular color.

ADDITIONAL MATERIALS NEEDED

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

- 1 lamp or bright window, not in direct sunlight
- 1 15 x 15-cm piece of clear, colorless transparency film or report cover
- several 15 x 7.5-cm pieces of cellophane, colored transparency film, or report covers
- 3 rubber bands

HYPOTHESIS GENERATION

Question

Are snails able to detect different colors?

Sample Hypothesis

Snails will show no preference for light of a particular color.

Rationale

Although it is not likely that an animal which is active at night is able to discern color, it is possible that it can detect differences in wavelength characteristic of dawn and twilight.

Sample Experimental Procedure

1. Determine the number of sheets of each colored transparency required to transmit equal light intensities as follows:
   A. Arrange the light meter so that the needle is not deflected full scale.
   B. Set a single sheet of the darkest color over the sensor and record the result.
   C. Remove that sheet and replace it with a single sheet of another color. Add more sheets of the less light-blocking color until the meter reading is the same as the value you recorded for the single darker sheet. Use the same number of sheets of the second color in your experiment.

D. Continue with this technique until you have determined the number of sheets needed for each color to transmit equal light intensities.

2. Then, repeat Steps 1 to 4 of the Sample Experimental Procedure in Variation 3.
3. Quickly roll colored transparent film, instead of black construction paper, over one half of the tube and fasten it with a rubber band. See Figure 13.

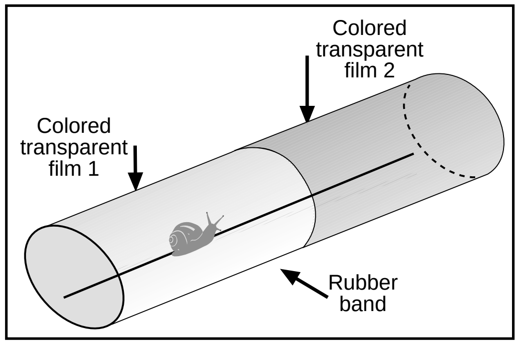


Figure 13. Cover the transparency with colored transparent film.

4. Cover the other half of the tube in the same manner, but use a different color. Table 10 shows the arrangement of colors you should create.

Table 10. A grid of color combinations providing an equal number of chances to choose each color.

	Color A	Color B	Color C	Color D	Color E
Color A	XXX	AB	AC	AD	AE
Color B	XXX	XXX	BC	BD	BE
Color C	XXX	XXX	XXX	CD	CE
Color D	XXX	XXX	XXX	XXX	DE
Color E	XXX	XXX	XXX	XXX	XXX

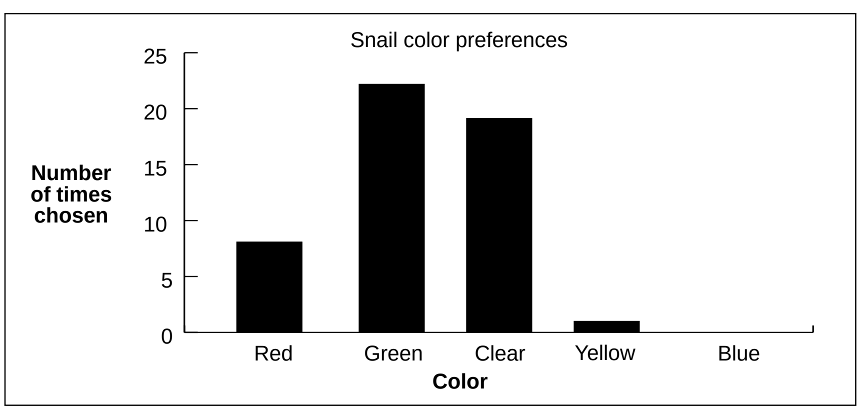
5. Lay the tube under the lights. Do not put it close to an incandescent bulb because the temperature can become dangerously high for the snail.
6. Record the color the snail selected at 1-minute intervals over a 10-minute time period.
7. Use a damp paper towel to clean slime from the snail side of the transparency film and repeat your observations with another snail.

SAMPLE DATA ANALYSIS AND INTERPRETATION

Sample Data

Table 11. Snail color preferences.

n=10	Red	Green	Clear	Yellow	Blue
Number of times chosen	8	22	19	1	0



Graph F. Snail color preferences.

TEACHING TIPS

- Once your students have rolled the colored transparency films to the correct size and fastened them with rubber bands, they may be able to slip them on and off the colorless tube for quick setup. Alternatively, you may want to provide tape for fastening the films.
- Remember that if you want to do a Chi-Square Goodness-of-Fit, you must have expected values of at least five for each snail.
- This setup is more a technique that detects unsatisfactory light environments rather than preferred light environments. It is a good starting point that can lead to some interesting discussions.
- Keep a record of the snail responses in the design grid, then summarize the data as shown in Sample Data of this variation.
- Encourage your students to graph their data with the colors arranged by increasing wavelength: violet-blue-green-yellow-orange-red.
- Transparent colored films, such as cellophane or report covers, are not spectrally pure. This will not invalidate the results if you state them in terms of the materials used rather than specific wavelengths.
- The intensity of light transmitted by a single sheet of different colors will be different. To avoid complicating the experiment, adjust the number of sheets you use to create the different colors to pass light of approximately the same intensity. Your adjustment will be approximate because you can make only whole-sheet adjustments.
- An inexpensive plant light meter

(Continued on p. 295)

Interpretation

If snails do not exhibit color preference, we may not assume that they do not have color vision; only that over the range tested, they had no preference. Reject the hypothesis if any color attracts more snails than expected by chance alone. The color with the most snails is the preferred color. Accept the hypothesis if an equal number of snails moved toward each color.

Analyze the data statistically with Chi-Square Goodness-of-Fit. To perform the statistical test:

1. *Determine expected frequencies*: You expect a particular relationship in your results. The relationship in this example is the same number of individuals in each treatment or 50/5 = 10. That is, the total number of observations divided by the ways they can be observed.
2. *Calculate Chi-Square*: For each color, subtract the expected value from the observed value, square the difference, and divide by the expected value. In equation form, this is:

$$\sum = \frac{(\text{Observed - Expected})^2}{\text{Expected}}$$

   Add all of the quotients to obtain a Chi-Square value.
3. *Compare*: In row (v) of the table, use one less than the number of classes. In this experiment, we have used five classes. Compare your calculated Chi-Square with the values in the table for $v = 4$. For $\alpha = 0.05$, Table 12 shows the expected Chi-Square values.
4. *Conclude*: If your calculated value is greater than the value in the table, you may conclude that the difference between your results and those hypothesized is not likely due to chance alone. You, therefore, reject the null hypothesis. With an $\alpha = 0.05$, 5% of the time you may reject a hypothesis of no difference when a difference exists.

TEST QUESTION

Could color play a role in food location by your snails?

SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

Blind or Visually Impaired

- Use a light sensor to compare transmission of equal light intensities through sheets of colored transparent material. In order to test more than one color at a time and to have better conditions for using the light probe to follow the travels of the snail, a simple device is suggested.
- Place strips of colored transparent material on the sides and lid of a clear, plastic shoe box 30 to 35 cm (12 to 14 inches). Drape a black cloth over the end.
- The lid on the box can be removed easily to use the light sensor. If the lid that comes with the box is a solid color, substitute a clear piece of plastic for the cover.
- See Figure 14.

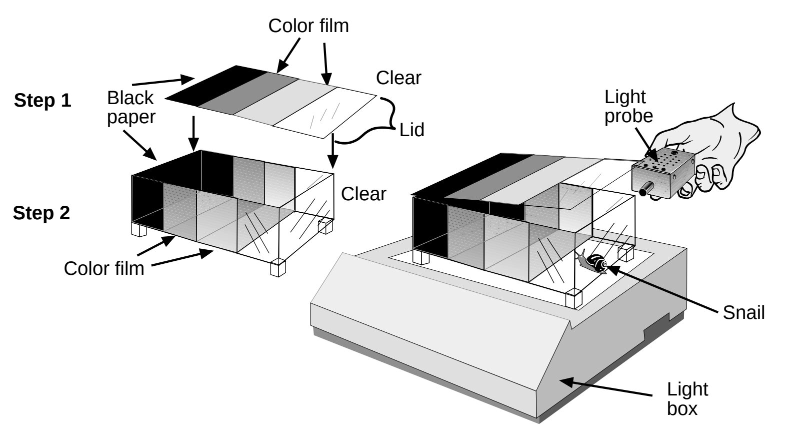


Figure 14. Simple device for testing the effects of more than one color at a time

Deaf or Hard-of-Hearing

- Be aware that Chi-Square may be new to most of the students and may be difficult to understand for some students who are visually or hearing impaired. This may require the use of a magnetic board and large magnetic numbers. The assistance of math teachers and mature lab assistants may be beneficial.

TEACHING TIPS

with a linear scale should be adequate to measure the light transmitted. Biological supply companies, discount mail-order tool catalogs, and nurseries usually sell these devices. You can make a light meter with the solar cell from an inexpensive calculator and a voltmeter. You may be able to borrow a voltmeter from the physics or electronics instructor or from the maintenance shop.

- If a window with good light is not available, an electric viewing box, such as that used to sort 35 mm slides, is suggested. The school's Audio Visual (AV) Department may have a 35mm viewing box.

Table 12. Expected Chi-Square values for $\alpha = 0.05$.

v	Expected Chi-Square value
1	0.455
2	1.386
3	2.366
4	3.357
5	4.351
6	5.348

Answer to Test Question

If snails showed a strong preference for particular colors commonly found in plant leaves and fruits, these colors may help the snail locate food.

TEACHING TIPS

- Heat transfer will be better through the transparency film than through the glass plate.
- Meat or pastry trays are good sources of Styrofoam™ for insulation.
- You may want to modify the design by placing four bags together and setting the snail at the intersection of the bags. See Figure 15. If you use this modification, select only four temperatures to test. This modification will require fewer replicates.

Figure 15. Setup with four bags at varying temperatures.

- The movement of poikilotherms, including snails, is influenced strongly by temperature. *Limax* is a slug with only a rudimentary shell. For *Limax maximus*, both the speed that a muscle wave moves along the foot and the step length (the distance a point on the foot moves as the wave passes over it) affect the rate that the snail moves. An increase in temperature increases the wave frequency, but decreases the step length. Over the temperature range of 6 to 30˚C for this species, the rate of movement increases to 15˚C, then decreases as the temperature increases (Jones, 1975).
- To have expected values of at least five for each cell, remember Chi-Square Goodness-of-Fit to determine the number of trials and the number of temperatures to test.
- This experiment is more a technique that detects unsatisfactory temperature environments rather than preferred temperature environments. It is a good starting point for interesting discussions.
- Keep a record of the temperature choices using the original design grid. Summarize the data as presented in the Sample Data. Refer to the original data record as well as to the summary when writing the conclusions and discussion.

VARIATION 5

The Effect of Temperature on Snail Behavior

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 determine if snails show positive or negative thermotactic behavior.

ADDITIONAL MATERIALS NEEDED

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

- 2 zip-closure, plastic sandwich bags
- 1 centigrade thermometer
- 2 600-mL beakers or 16-oz jars
- ice
- 600 mL tap water
- 1 Styrofoam™ meat or pastry tray
- 1 15 x 15-cm piece of transparency film or report cover

HYPOTHESIS GENERATION

Question

Do snails have a temperature preference?

Sample Hypothesis

Snails will have no temperature preference.

Rationale

The phrasing is designed to use statistical analysis. One would expect to find some temperature preference. You might even expect that preference to be narrow in snails who regulate their body temperature behaviorally, rather than metabolically.

Sample Experimental Procedure

1. Use ice and tap water to create two beakers of 300 mL of water at two different temperatures. Temperatures of 5 to 30˚C should be safe for the snail. Table 13 shows a possible range of temperatures.
2. Put the water from each beaker into a sandwich bag. Express the excess air and seal the bag.
3. Put the bags next to one another and set your glass plate or transparency on top of them. See Figure 16.
4. Put your snail on the glass plate or transparency so that its body is at the juncture of the bags and parallel to it.
5. Lightly mist the plate or transparency and allow the snail time to move and record the temperature it selects.
6. Use a damp paper towel to clean slime from the snail side of the glass plate or transparency film, and repeat your observations with another snail.

Table 13. A grid of temperature combinations providing an equal number of chances to choose each temperature.

	50˚C	40˚C	25˚C	10˚C	0˚C
50˚C	XXX	AB	AC	AD	AE
40˚C	XXX	XXX	BC	BD	BE
25˚C	XXX	XXX	XXX	CD	CE
10˚C	XXX	XXX	XXX	XXX	DE
0˚C	XXX	XXX	XXX	XXX	XXX

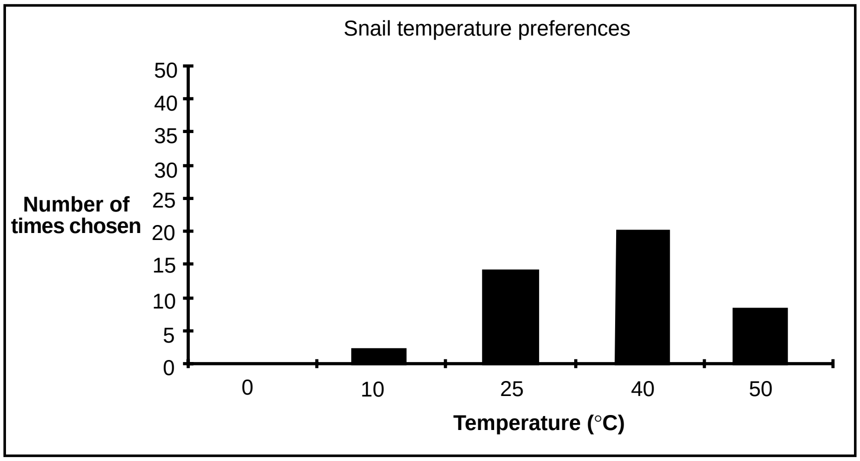
Figure 16. Setup for experiment.

SAMPLE DATA ANALYSIS AND INTERPRETATION

Sample Data

Table 14. Snail temperature preferences.

	50˚C	40˚C	25˚C	10˚C	0˚C
Number of times chosen	8	17	14	1	0



Graph G. Effect of temperature on snail behavior.

TEST QUESTION

Explain why snails are most active at night or on cloudy days, but not active during cold winters.

SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

Blind or Visually Impaired

- This experiment requires no new laboratory techniques for the students who are visually impaired. They may wish to use the Talking Thermometer to check the temperature on top of the glass plate in various locations.
- See Considerations for Variation 4 for Deaf or Hard-of-Hearing.

Interpretation

Reject the hypothesis if any temperature has more snails than expected by chance alone. The temperature with the most snails is the preferred temperature. Accept the hypothesis if an equal number of snails moved into each area.

Answer to Test Question

The snails never chose the highest temperature offered. They were most active at cooler temperatures like those that occur at night during the summer or on cloudy days. Even if they could find food on a very cold winter day, they only chose the coldest temperature when the other choice was a high temperature.

VARIATION 6

The Effect of Plant Food Choice on Snail Feeding Behavior

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 snails have a preference for a specific plant food.

ADDITIONAL MATERIAL NEEDED

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

- pieces of several different types of food, including apples, lettuce, spinach, broccoli, and carrots.
- 1 balance *(optional)*
- 1 blender or mortar and pestle *(optional)*

HYPOTHESIS GENERATION

Question

Do snails avoid food that is toxic?

Sample Hypothesis

When offered a choice of fresh produce, snails will show no preference.

Rationale

Snails must be able to distinguish between foods that are toxic and those that are safe.

Sample Experimental Procedure

1. Prepare the filter paper strips with extracts of the various foods as in the Core Experiment.
2. Place the strips of paper in a circle on the plastic sheet protector leaving an equal space between each piece. See Figure 17.
3. Lightly mist or dampen the plastic inside the circle.
4. Place one snail in the middle of the circle.
5. Observe and record which direction it travels and how it reacts when it reaches each strip.
6. If the snail does not move after it has reached a strip, pick it up carefully and return it to the middle so it is facing another direction.
7. Repeat Steps 4 to 6 four times.

Figure 17. Arrangement of plant food choices.

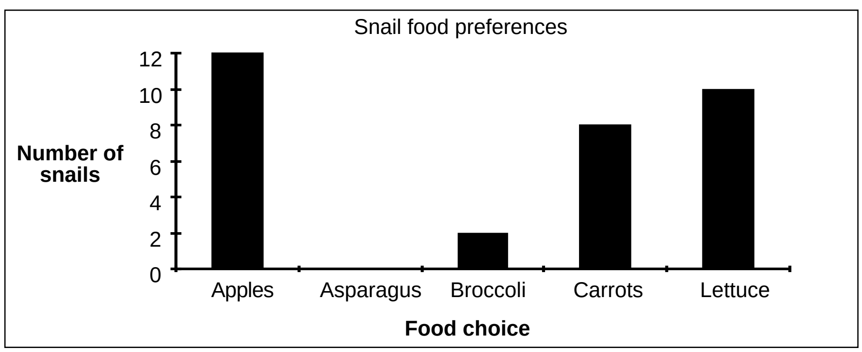
SAMPLE DATA ANALYSIS AND INTERPRETATION

Sample Data

Table 15. Snail food preferences.

	Food choice				
	Apples	Asparagus	Broccoli	Carrots	Lettuce
Number of snails selecting the food	12	0	2	8	10

If you want to do a statistical analysis, Chi-Square Goodness-of-Fit is appropriate (see Variation 4).



Graph H. Snail food preferences.

TEST QUESTION

If you were keeping snails in a terrarium, what would you feed them to be certain that they had the essential nutrients for growth and health?

TEACHING TIPS

- The snails may not respond well to this design if they are not hungry. Students may want to do this as an extended exercise, collecting information on two foods at a time and overnight. If a snail does not eat a particular food, it may be that the uneaten food was not encountered at all or was encountered when the snail was no longer hungry.
- If students have fed the snails lettuce during the introductory period of this lab and recorded the amount eaten, they will have some idea of how much a single snail will eat. They can then offer two different foods, each in half the amount that a snail would normally eat. They could offer four foods at once, each in one-fourth the amount of the total food normally consumed, and so on.
- A good way to measure the amount eaten is to mass the fresh food when it is offered and again when it is removed. A separate container with wet paper towel and food pieces the same size as those offered can be used to adjust the final mass for loss by evaporation.
- A third possible design for this variation is to use the puree without the paper.
- In the field, snails are selective in their diet and may not eat from all their preferred plants in an area (Runham, 1975). They may have short term preferences, eating one type of food exclusively for several days. The maturity of the plant material may alter its palatability.
- Juvenile and mature snails differ in their food preferences (Runham, 1975).

Interpretation

Reject the hypothesis if any food is selected more or less often than the others. Accept the hypothesis if an equal number of snails select each food choice.

Answer to Test Question

Although broccoli and asparagus are nutritious for humans, they would not be good choices to feed snails. Snails do not eat them. Terrarium snails would be fed a mixture of fresh vegetables including apples, lettuce, and carrots. These foods were most often chosen by the experimental animals.

SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

Blind or Visually Impaired

- Notch the edge of the large filter paper to identify the food at each location. The samples may be placed on a large circle of wax paper taped to the lab table. Some students who are visually impaired will be able to identify the pureed foods by smell.

VARIATION 7

The Effect of Bacterial Decomposition on Food Preference by Snails

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 if fresh or decaying lettuce is preferable to snails.

ADDITIONAL MATERIALS NEEDED

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

- decayed lettuce
- fresh lettuce

HYPOTHESIS GENERATION

Question

Do snails prefer fresh or decayed food?

Sample Hypothesis

Snails will prefer fresh lettuce over decaying lettuce.

Rationale

Creation of silage makes more protein available to livestock. Spoilage of the lettuce similarly may make different nutrients available to the snails.

Sample Experimental Procedure

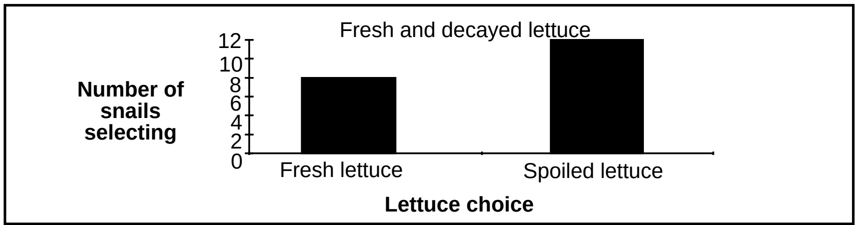
1. Prepare the separate filter paper halves with purees of the fresh or decayed lettuce and place them in the glass dish as shown in the Core Experiment.
2. Lightly mist or dampen the plastic between the filter paper halves.
3. Place 1 snail between the filter paper halves as shown in the Core Experiment.
4. Observe and record which direction the snail moves and how it reacts when it reaches each half.
5. If the snail does not move after it has reached a half, pick the snail up carefully, return it to the center of the dish, and face it in the other direction.
6. Repeat Steps 3 to 5 four times.

SAMPLE DATA ANALYSIS AND INTERPRETATION

Sample Data

Table 16. Snail food preferences.

	Food choice	
	Fresh lettuce	Decayed lettuce
Number of snails selecting the food	8	12



Graph I. Snail food preferences.

TEACHING TIPS

- To decay the lettuce slightly, store it wet in plastic in the refrigerator for several days to encourage bacterial growth.
- Students may volunteer to bring lettuce from home.
- Several snails have been shown to prefer decaying vegetation to fresh vegetation in the field (Runham, 1975).

Interpretation

Reject the hypothesis if the same number of snails select each choice, or if more snails select the decaying lettuce than the fresh lettuce. Accept the hypothesis if more snails choose fresh lettuce.

Answer to Test Question

Straw contains few nutrients and may not be very attractive to snails. The experimental animals expressed a strong preference for decaying lettuce. They may be attracted strongly to this garden plot and consume the crop when the decayed lettuce is exhausted. The lettuce-mulched plot may be predicted to have the greatest damage. However, if the snails cannot move between plots, the greatest damage will occur in the straw-mulched plot because snails do eat fresh vegetables.

TEACHING TIPS

- The recovery of animal material from the gut of field-collected snails does not seem to be the result of accidental ingestion. More than 20% of the specimens of one grassland species of herbivorous snail contained animal parts in their guts (Runham, 1975). These parts included both segmented worms and arthropods.
- *Lymnaea stagnalis*, the great pond snail, although primarily an herbivore, consumes dead animals. It has an ability to detect animal material at a distance, but not plant material (Runham, 1975).
- Snails may be omnivorous, but most are herbivorous.
- Although you could puree the materials for this exercise, the field snails will probably not encounter the food processed so completely. A "stepped-on colleague" is more likely to be the animal food encountered.
- *Helix pomatia* hatchlings devour unhatched eggs (Cain, 1983) and several other species of snails act as predators on occasion.
- Remember that if you want to do a Chi-Square Goodness-of-Fit, have expected values of at least 5 for each cell. Use this desired expectation to determine the number of trials and temperatures you will test. You will want to combine Lettuce 1 and Lettuce 2 for the statistical analysis. Since the snails have an equal opportunity of choosing one of four stations in the sample below, the expected value for lettuce should be half the number of snails observed.

TEST QUESTION

Would you predict that more crop damage will occur in a small garden plot where plants are heavily mulched with waste lettuce leaves or in a plot with the same distribution and kinds of plants but with a mulch of nutrient-poor straw? Explain your prediction.

SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

- See Considerations for Variation 6.

VARIATION 8

The Effect of Plant Versus Animal Food Choice by Snails

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 determine whether *Helix* prefers animal or plant material for food.

ADDITIONAL MATERIALS NEEDED

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

- a selection of animal materials such as dead snails, sections of dead earthworms, hamburger, or other supermarket raw meats

HYPOTHESIS GENERATION

Question

Will snails prefer plant material over animal material?

Sample Hypothesis

Snails will prefer plant material when offered a choice of plant or animal material.

Rationale

Helix is such a serious agricultural pest that there are restrictions on importing it into some states.

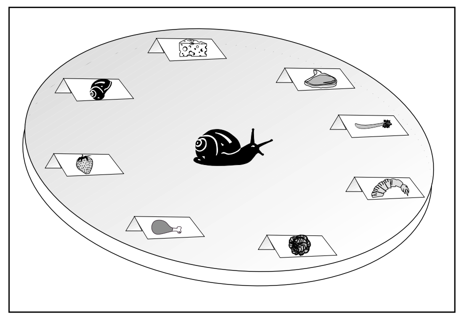


Figure 18. Arrangement of plant and animal food choices.

Sample Experimental Procedure

1. Arrange plant and animal food choices in a circle on the glass or plastic experimental surface leaving an equal space between each choice. Be certain to provide an equal number of plant and animal choices.
2. Lightly mash each food choice to simulate damage that might occur in the field.
3. Lightly mist or dampen the plastic inside the circle.
4. Place one snail in the middle of the circle.
5. Observe and record which direction it travels and how it reacts when it reaches each food choice.
6. If the snail does not move after it has reached a choice, pick it up carefully and return it to the middle so it is facing another direction.
7. Repeat Steps 4 to 6 several times.

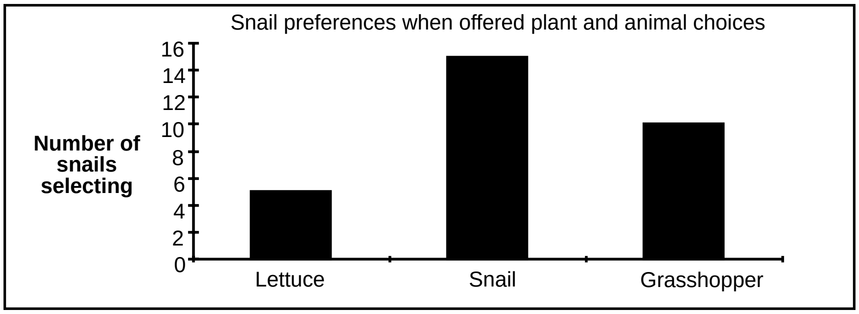
SAMPLE DATA ANALYSIS AND INTERPRETATION

Sample Data

Table 17. Snail preferences from plant and animal food choices.

	Food choice			
	Lettuce 1	Lettuce 2	Snail	Grasshopper
Number of snails choosing a particular food source	2	3	15	10

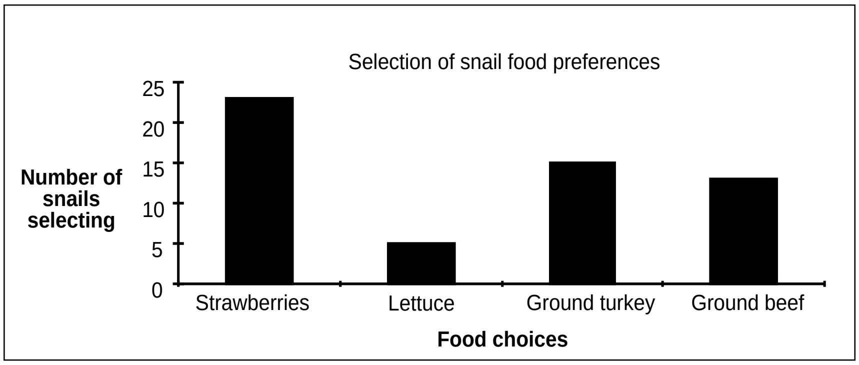
A Chi-Square Goodness-of-Fit analysis is appropriate for this design (see Variation 4).



Graph J. Snail preferences for plant and animal food choices.

TEST QUESTION

Graph K represents the results of an experiment testing snail food preference. Do these snails show a preference for any particular food? If so, which food do they prefer? How do these results compare with those you obtained?



Graph K. Snail preferences for 2 plant and 2 animal food choices.

SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

Blind or Visually Impaired

- Offer the snail two circles of paper with food choices to allow room between the foods. Make a third run with the best choices on other papers to determine final choices.
- See Variation 4 for additional modifications.

Deaf or Hard-of-Hearing

- See Variation 4.

VARIATION 9

The Effect of Mechanical Disturbance on Snail Activity

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 perturb snails gently with shell percussion and shaking to determine if these disturbances influence behavior.

ADDITIONAL MATERIAL NEEDED

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

- 1 8-cm cotton swab
- 1 clock with second hand

HYPOTHESIS GENERATION

Question

Will snails respond to mechanical stimulation?

Interpretation

Reject the hypothesis if the same number of animals select the plant and animal foods or if more snails select the snail and grasshopper than select plant material. Accept the hypothesis if more snails select lettuce than select snail and grasshopper combined.

Answer to Test Question

Snails show a preference for strawberries and animal foods. Here, only half the snails chose animal foods.

TEACHING TIPS

- Some researchers (Kerkut & Walker, 1975) have observed snails withdraw in response to mechanical vibration, but others (Machin, 1975) working with hibernating snails have observed activation by percussion, shaking, or rotation.
- Snails can habituate to a stimulus (Kerkut & Walker, 1975), so do not stimulate the same snail in the same way more than 20 times.
- Turn off the lights in the classroom and increase the relative humidity in the vicinity of the snails for this activity. If you started with estivating or hibernating snails, they will be more likely to continue an initiated arousal if you provide conditions under which they would be active normally.
- Expect active snails to move 2 to 10 cm per minute (Jones, 1975).
- A team of four students can collect data quickly on this experiment if they work with a dozen snails. Three students could deliver the percussive stimulus, while the fourth student gently shakes three snails on the same plastic sheet.
- Try this with your unresponsive snails. If you still have trouble arousing them, lower their temperature to about 10˚C and increase their relative humidity to 100% for four hours. This activated more than half the snails in one experiment with 48 animals (Machin, 1975).

Sample Hypothesis

Snails will exhibit defensive withdrawal behavior if they are disturbed mechanically.

Rationale

Turtles have a shell and often withdraw into it if disturbed. Disturbances, such as a tap, may be the initial encounter with a hungry mouse, bird, frog, toad, lizard, or salamander. Hiding in the shell would be a better strategy than outrunning one of these predators.

Sample Experimental Procedure

1. Place a snail on a glass plate or plastic sheet and mist the surface lightly with spring water.
2. Provide no further stimulation to 1 snail.
3. Stimulate each of the remaining 2 snails continuously for 2 minutes using a different stimulation technique for each snail as follows:
   A. For percussive stimulation, rest a cotton swab midway between 2 fingers with one tip of the swab just resting on the back of the snail's shell. *Gently*, flick the free end downward about 10 times per minute. See Figure 19.
   B. For shaking stimulation, lift the plastic sheet off the lab bench and move it *gently* left to right and back through a displacement of about 15 cm, then pause. Repeat this process at the rate of 10 complete displacements per minute.

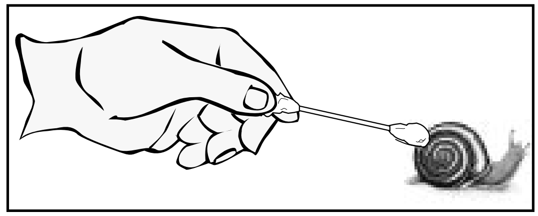


Figure 19. Procedure for using a cotton swab as a percussive stimulation.

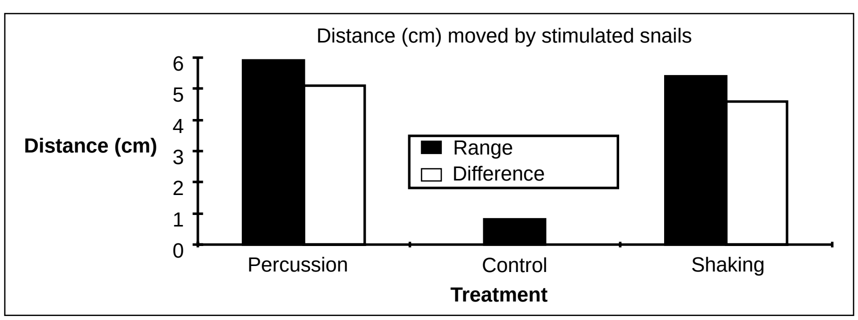
4. As soon as you have finished stimulating your snail, place a piece of lettuce or the preferred food 10 cm from its anterior end. Record the rate of movement toward the food. Continue your observation for 5 minutes, moving the lettuce just beyond the reach of the snail if necessary.

SAMPLE DATA ANALYSIS AND INTERPRETATION

Sample Data

Table 18. Distance moved and behavior exhibited by mechanically stimulated snails.

	Distance moved with treatment		
Trial	None (cm)	Percussion (cm)	Shaking (cm)
1	0 No activity	10.0 Withdrew, then emerged and moved quickly	3.4 Did not withdraw
2	0 No activity	2.3 Withdrew, then emerged and moved, but stopped after a short distance	5.2 Did not withdraw
3	2.3 Moved toward lettuce	5.5 Withdrew, then emerged and moved slowly	7.5 Did not withdraw
Mean	0.8	5.9	5.4



Graph L. Effect of mechanical stimulation on snails.

TEACHER'S NOTES

Interpretation

Reject the hypothesis if some snails withdrew and some did not, and discuss what was observed. The snails responded differently to the two kinds of mechanical stimulation. Accept the hypothesis if stimulated snails withdrew into their shells.

TEST QUESTION

What natural changes in the environment might result in mechanical stimulation? What snail responses to these stimuli might be adaptive in the field?

SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

Blind or Visually Impaired

- Have students create mechanical vibration and other disturbances for the snails. The braille watch can be used as a timer.
- Tape braille letters to the shell of each snail as a label when more than one snail is being used. Drops of glue also may be used as a marking device on the shells.

VARIATION 10

The Effect of Surface Texture on Snail Movement

Note to Teachers: In addition to the information found in the Core Experiment, the following material has been provided for Variation 10.

SYNOPSIS

Students will determine whether snails move at the same rate over smooth and coarse surfaces.

ADDITIONAL MATERIALS NEEDED

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

- waterproof sandpaper of various grit sizes
- 1 tape measure or ruler and string
- 1 clock or watch with second hand

HYPOTHESIS GENERATION

Question

How does the texture of a surface affect snail movement?

Sample Hypothesis

Snails will move more slowly over a rough surface than over a smooth surface.

Rationale

A smooth surface should provide better contact with the mucus and cilia that are involved with the snail muscles that create movement.

Sample Experimental Procedure

1. Mist each selected sandpaper with spring water to create a wet surface without standing water. You may blot the surface with paper towels to remove excess water.
2. Place a snail on the sandpaper 5 cm from a preferred food.
3. Record the time required for the snail to reach the food.
4. Calculate the rate of movement in centimeters per minute.

Answer to Test Question

Mechanical stimulation could simulate the movement of vegetation at nightfall or from an arriving storm. It might be the tap of a rain drop. If these changes are initiated by the stimulation, it would be advantageous for the snail to increase its activity of foraging for food with little risk of dehydration. However, if the mechanical stimulation is the result of a predator testing the shell for contents, it would be advantageous for the snail to withdraw. It is unlikely that it can outrun a predator.

TEACHING TIPS

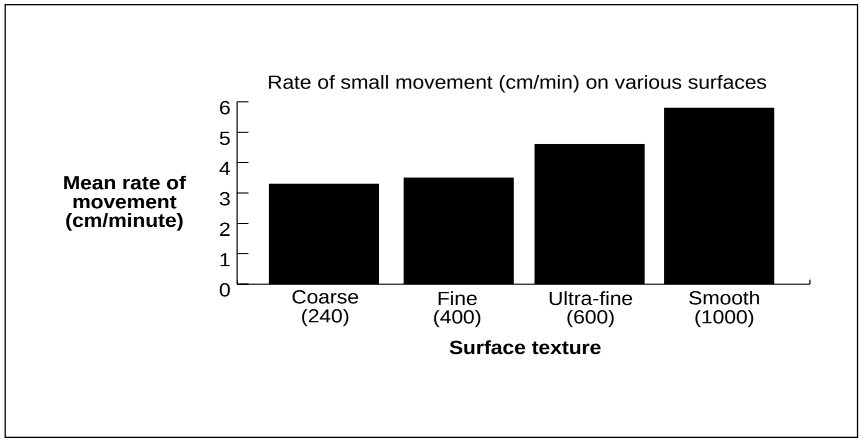
- Waterproof sandpaper is not made in extremely coarse grits, but other sandpaper tends to come apart when wet.
- You are more likely to find waterproof sandpaper in a variety of grits at an automobile parts supply store than at a hardware store.
- If the snail does not travel in a straight line, you may measure how far it went by laying a string along its route and then measuring the string.
- The main adhesion between a land snail and the surface that it moves over is the mucus distributed over the foot surface by cilia. The mucus appears to be necessary for movement (Jones, 1975).
- Within a species, small individuals move faster than large individuals (Hyman, 1967). *Helix aspersa* moves about 7.5 cm/minute, and *Helix pomatia* moves from 6.25 to 7.8 cm/minute.

SAMPLE DATA ANALYSIS AND INTERPRETATION

Sample Data

Table 19. Rate of movement (cm/minute) for snails on various surfaces.

Trial	Smooth (glass) cm/minute	Ultra-fine grain (600) cm/minute	Fine grain (400) cm/minute	Coarse grain (240) cm/minute
1	5.2	5.0	4.5	4.2
2	5.0	5.1	4.0	2.0
3	7.1	3.8	2.0	3.8
Mean	4.4	4.7	3.6	3.3



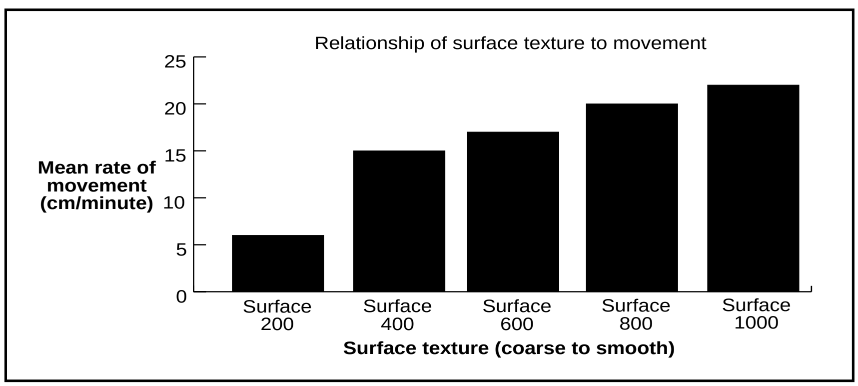
Graph M. Influence of surface texture on the rate of snail movement.

Interpretation

Reject the hypothesis if the average rate of movement on any textured surface is greater than that on the glass surface. Accept the hypothesis if the slope of the line is negative.

TEST QUESTION

Study the graph of snail movement in Graph N. Compare the rate of movement of these snails over various surfaces. Is the performance of these snails comparable to that of your experimental animals?



Graph N. Influence of surface texture on the rate of snail movement.

Answer to Test Question

The smoother the surface, the faster these snails are able to move. Our snails showed the same pattern of change in rate, but they moved much more slowly.

SUGGESTED MODIFICATIONS FOR STUDENTS WHO ARE EXCEPTIONAL

Blind or Visually Impaired

- Provide braille timing devices with a second hand if available. The class timer used in science labs is usually sturdy enough to be used manually by the student who is blind.
- Provide braille tape measures and rulers.
- Use braille graph paper for a line graph. The points are marked on the paper and are connected with a French curve.

A Study of Snail Behavior

Directions for Students

INTRODUCTION

Have you ever observed animals in action? Bees moving towards a flower, ants toward food, a snail towards a leaf, or a roach darting for cover when a light is turned on? Why are these animals exhibiting these behaviors? How do they "know" where to go? Is it instinct? Have they learned these behaviors?

Most of us are aware of the senses of touch, taste, smell, seeing, and hearing. Which sense do you use the most? Which of your senses is the most reliable? Do other animals also have these senses? In this activity your teacher will provide you with a snail to observe. Spend 5 to 10 minutes watching the snail. As you watch your snail, how could you test whether a snail is able to sense its environment?

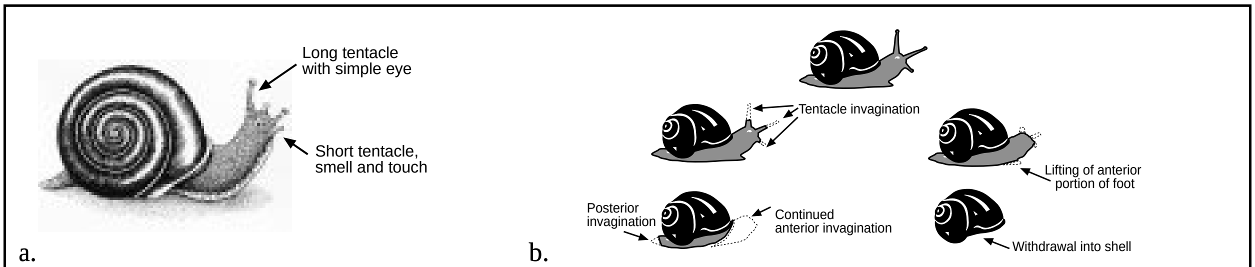


Figure 1. a. Snail with tentacles' functions identified. b. Normal retraction sequence in snails.

OBJECTIVES

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

- Determine how snails respond to chemical stimuli, including chemicals found in their environment.
- Discuss why snails may respond differently to various chemical stimuli in their environment in terms of evolution.

MATERIALS NEEDED

For each team of two students, you will need the following:

- 1 1-L plastic or glass jar with lid or 1 3 x 10 x 10-cm plastic sandwich container with snap-on lid
- 1 8 x 8-cm piece of 2-cm thick Styrofoam™
- 1 compass with pencil
- 2 25-cm filter paper pieces, paper towels, or coffee filters
- 1 mister/sprayer
- 1 large land snail (*Helix)*
- 1 20 x 30-cm plastic sheet protector, report cover, or glass plate
- 1 25-cm diameter glass pie plate or clear plastic dish
- 1 permanent marker
- 1 metric ruler
- 1 pair of scissors
- 10 mL spring water
- 3 25-mL vials
- 1 forceps
- 1 clock or watch with second hand
- 10 mL 10% spinach extract
- 10 mL 10% glucose solution
- 1 lab journal

SAFETY PROCEDURES

- Use only substances that can be poured safely down the sink to make disposal easier.
- Do not use chemicals that are toxic or dangerous to humans.
- Never use pure caffeine or nicotine. Avoid pesticides and insecticides.
- Do not release snails into gardens or greenhouses. Both *Helix aspersa* and *Helix pomatia* are European natives that are serious agricultural pests.
- Do not permit snails to crawl on lab benches.
- Wear safety goggles and aprons when working with chemicals.
- Students should wash their hands before and after working with the snails.
- Keep hands and fingers away from the compass point when punching holes in lids.
- Handle snails with care. Never pull or lift the animal directly from a surface. Rather, gently slide the animal from a surface.
- Always maintain a moist environment. Clean container at least once a week. Do not use distilled water or tapwater directly on the snails.

STUDENT LITERATURE SEARCH SUMMARY WITH REFERENCES

Do a literature search on the topic of snails and chemical sensitivity. Summarize your findings and cite your references. 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. Why is controlling variables important?
4. What is the variable in your experiment?
5. How will you control other potential variables?

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 must have your teacher approve this protocol before 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 bar graph showing the number of snails responding positively and negatively to each chemical tested.
2. Do your data support or refute your hypothesis? Explain.
3. In one set of student experiments, 100% of the snails showed a positive response to spinach; while in another set of student experiments, 75% of the animals showed a negative response. What explanations might account for the difference?
4. Would knowing that the sample size in one experiment was 30 snails, while in the other experiment it was 4 snails, change your confidence in the results? Explain.
5. How could you test whether the aroma of the material influences the movement of a snail toward it?

DESIGN OF VARIATIONS OF CORE EXPERIMENT

After collecting and analyzing the data from the Core Experiment and sharing the results and conclusions with the class, brainstorm ideas for experiments you could do next. Think of questions that occurred to you as you conducted the Core Experiment on snails and environmental factors. What quantifiable experiments could be done based on the observations you made? Questions other students have studied include the following:

- How will changing the solution's concentration affect the snail's response?
- What type of geotactic response do snails exhibit?
- What form of phototactic response do snails exhibit?
- Are snails able to detect different colors?
- Do snails have a temperature preference?
- Do snails avoid food that is toxic?
- Do snails prefer fresh or decayed food?
- Will snails prefer plant material over animal material?
- Will snails respond to mechanical stimulation?
- How does the texture of a surface affect snail movement?