THE NEURO-MUSCULAR BASIS OF EARTHWORM MOVEMENTS: EFFECTS OF PHYSICAL AND CHEMICAL ENVIRONMENTAL AGENTS

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TABLE OF CONTENTS

SECTION 1 — INTRODUCTION
RATIONALE AND GOAL 4
LEARNING OBJECTIVES 6

SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS
OVERVIEW 9
INSTRUCTIONAL COMPONENTS 9
PLANNING TIMETABLE 10
ADVANCED PREPARATION GUIDE 13
DAILY LESSON PLANS 17

SECTION 3 — SUPPORT MATERIALS
NATIONAL SCIENCE STANDARDS 148
INSTRUCTIONAL ACTIVITIES – STUDENT COPY MASTERS 154
MATERIALS AND POSSIBLE SOURCES 234
SOLUTION PREPARATION INSTRUCTIONS 235
SECTION 1 — INTRODUCTION

RATIONALE AND GOAL
LEARNING OBJECTIVES
   AFFECTIVE OBJECTIVES
   CONTENT OBJECTIVES
SKILLS
KNOWLEDGE
RATIONAL AND GOAL

RATIONALE

This module is designed to incorporate environmental and human health issues in the basic science content of the high school level curriculum in an engaging manner. It provides age-appropriate activities for high school level learners and readily-used support materials for educators.

Environmental sciences – particularly those related to human environmental health – provide students with a cross-disciplinary focus for science learning that has personal and local relevance as well as developmentally appropriate content. Students at this stage are generally quite interested in understanding systems and relationships. They can begin to recognize the connection between explanations and evidence and become very engaged in exploring that connection. Because they often seek to confirm their personal beliefs and concepts, they can be challenged and informed by addressing controversial topics.

The module features serve student interests by:

- using a familiar, live species for understanding locomotion and exploring possible effects of environmental agents,
- having students select the environmental agents to be tested,
- having students design their own experiment,
- focusing on a nerve-muscle interaction similar to one in humans,
- exploring a possible link to human health, and
- providing activities that cross curricula and tap multiple intelligences.

Finally, the module is designed for ready use by the teacher and student. It consists of anticipatory, laboratory, and reflective activities designed to generate critical thinking about neuro-muscular processes and environmental pollutants that affect them, and concludes with the question, “Might humans experience effects of environmental agents like those seen in exposed earthworms?”

GOAL

The goal of this module is to lead students to understand that substances harmful to human health are found in both natural environments and environments altered by humans. Understanding the consequences of exposure to these substances is the central focus. To achieve this goal, the module engages students with the three fundamental aspects of science learning: knowledge and application of basic concepts and principles, skills in scientific inquiry and processes, and understanding science as a human endeavor designed to address human problems and issues.
SECTION 1 — INTRODUCTION

Through the study of normal locomotion behaviors (crawling and simple reflexes) in earthworms and the impact of potentially toxic environmental agents on these behaviors, students will learn the basic concepts and principles of living organisms, including structure and function, regulation and behavior, and science and technology. They will gain and use skills in scientific processes and reasoning as they generate and clarify questions, conduct investigations, gather and analyze data, and make critical connections between evidence and explanation. They will also gain deeper insight into science as a human endeavor, one that serves to: a) understand environments, and b) characterize the impacts of human action on environments and of environments on human health.

In their study, students will articulate and refine their own broad questions about animal behaviors, their structural and functional basis, and their susceptibility to environmental agents. They will use appropriate tools and techniques to conduct controlled experiments using earthworms. They will gather and analyze data regarding the behavior of both control and experimental organisms. They will interpret these data to draw conclusions and generate explanations, predictions, and models of development. Finally, they will propose hypotheses that link their personal health to environmental agents and use those hypotheses to help them make personal and social decisions.
SECTION 1 — INTRODUCTION

LEARNING OBJECTIVES

AFFECTIVE OBJECTIVES
Through the implementation of the module students will develop an appreciation for life forms, their activities, and the environment. Specifically, they will:

- Appreciate that human activity in the environment may affect human health
- Appreciate the complexity of environmental health issues
- Identify with other life forms
- Appreciate the science process (questioning, designing and experiment, manipulating materials handling organisms, gathering and summarizing experimental evidence, and examining and re-examining ideas based on experimental evidence)

CONTENT OBJECTIVES
Additionally, students will learn the science related to normal and abnormal earthworm locomotion. Specifically, they will:

- Understand that a very simple-appearing earthworm is extremely well-adapted to its environment
- Understand basic external and internal anatomical structures of an earthworm and their relationship to its locomotion and survival
- Understand sensorimotor pathways and the simple reflexes they control in earthworms and humans
- Gather and analyze data on changes in simple reflexes in earthworms after contact with environmental contaminants
- Understand that abnormal locomotion and simple reflexes may at times be attributable to chemicals in the environment; namely, those naturally present and/or those introduced to the environment by humans
- Understand and summarize for earthworms and humans
  - Consequences of exposure to toxic chemicals
  - Ways to minimize exposure to toxic chemicals
- Apply the perspective and knowledge gained to other environments, organisms, and chemical agents
- Understand science as a process using direct experimentation
- Understand that an experimental conclusion is based on experimental data
- Use critical thinking to determine and explain whether data obtained permits a conclusion
- Understand that the need for a new hypothesis is based on experimental data
SECTION 1 — INTRODUCTION

SKILLS

- Caring for a pet/an experimental animal
- Using a data table
- Using a compound microscope
- Using websites
- Working cooperatively in a group

KNOWLEDGE

- Cell structure (particularly nerve cell) and processes (diffusion, osmosis)
- General classification (Kingdom, Phylum, Class, Order, Family, Genus, Species)
- Basic nerve function, especially synapse events
- Life science concepts (behavior, response, and enzyme)
- Some chemistry terminology (atom, element, ion)
- Human motion resulting from muscle acting on bone
- General ecology of the earthworm
  - Biotic and abiotic factors in an environment
  - Food web in a community
- Ecology of the earthworm
  - Characteristics of earthworms that render them sensitive to environmental agents
  - Biotic and abiotic factors in the worm’s environment
  - The earthworm’s place in the food web/food pyramid
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

OVERVIEW
INSTRUCTIONAL COMPONENTS
PLANNING TIMETABLE
ADVANCED PREPARATION GUIDE
DAILY LESSON PLANS
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

OVERVIEW

This module investigates earthworm locomotion and simple reflexes as well as the effect of aldicarb and other environmental agents on those behaviors. In a period of two to three weeks you will guide your students through the activities and concepts presented in the module. In that time they will observe the earthworm’s normal crawling, burrowing, and “startle response;” the anatomical bases for these movements; and the effect of both a highly toxic insecticide (aldicarb) and student-selected environmental agents on those movements. Instruction includes individual, small group, and whole class activities, some of which are inquiry-based.

The Planning Timetable below outlines advanced teacher preparation and classroom instruction. The Instructional Components section summarizes the concepts covered in the module, while the Daily Highlights outlines the materials and content preparation for instructional flow. The Detailed Instructional Guide provides comprehensive specifics on teacher preparation and class activities. Most instructional days will require an entire class period of 40 to 45 minutes. Although the schedule may be changed, the activities are best done in the conceptual progression provided.

INSTRUCTIONAL COMPONENTS

PART I – A MODEL FOR NORMAL LOCOMOTION (CLASS DAYS 1–2)

- Understanding Normal Earthworm Locomotion
- Examining the General Structural Basis of Locomotion

PART II – A MODEL FOR HUMAN HEALTH (CLASS DAYS 3-13)

- Understanding Normal Earthworm Responses to Threatening Stimuli
- Comparing Earthworm and Human Nervous Systems
- Examining the Lowest Observed Effect Concentration (LOEC) of Earthworm Sensory Neurons
- Articulating Possible Implications for Human Health

PART III – VIDEO EXTENSION AND GUIDELINES FOR STUDENT-DESIGNED EXPERIMENTS ON THE EFFECT OF SELECTED ENVIRONMENTAL AGENTS ON EARTHWORM LOCOMOTION (CLASS DAYS 14 & BEYOND)

- Observing the Effect of Aldicarb on Earthworm Burrowing
- Student-Designed Experiments (Optional)
- Investigating Earthworm Learning (Optional)
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

PLANNING TIMETABLE

The timetable below shows advanced teacher preparation and the classroom instruction schedule. Advanced teacher preparation takes place at least eight weeks before the module begins and includes determining student groups and having students bring in a washed and rinsed plastic gallon jugs. Once begun, the module calls for a minimum of eleven full days of classroom instruction. Suggestions for daily teacher preparation are provided in the Detailed Instructional Guide.

<table>
<thead>
<tr>
<th>TIME BEFORE</th>
<th>SUMMARY OF ACTIVITIES</th>
</tr>
</thead>
<tbody>
<tr>
<td>8 weeks or more</td>
<td>- Take materials inventory</td>
</tr>
<tr>
<td>14 days</td>
<td>- Determine student groups</td>
</tr>
<tr>
<td></td>
<td>- Have students bring CLEAN plastic gallon jugs</td>
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<tr>
<td></td>
<td>- Discuss substances that might be tested for an effect on worm movement</td>
</tr>
<tr>
<td></td>
<td>- Gain approval of school safety officer to use substances</td>
</tr>
</tbody>
</table>

PART I: A MODEL FOR NORMAL LOCOMOTION

Day 1

INTRODUCING AND BEGINNING THE MODULE

- Focus student attention on the subject of the first part of the module; namely, normal locomotion of earthworms, its structural basis, and using both as a model for human locomotion
- Elicit students’ prior knowledge about earthworms using “What Do I Know About Earthworms” (Class Instructional Aid, C#1)
- Have student groups read Student Activity Sheet “Earthworms!!!” (Student Activity, S#1), discuss, and check for understanding
- Prepare dechlorinated water
- Briefly review what the class learned about earthworms, and why the tap water was set out to become free of chlorine

Day 2

EXAMINING EARTHWORM ANATOMY AND NEURO-MUSCULAR INTERACTIONS

- Begin Lab “Earthworm Structures and the Work They Do” (S#2A/#2B)
## Part II: A Model for Human Health

### Days 3-4

**Observation of Normal Earthworm Responses to Threatening Environmental Stimuli**

- Finish “Earthworm Structures and the Work They Do” (S#2A/#2B)
- Discuss “Nerve-Muscle Interactions and Sensory Neurons” (C#2)
- Introduce and do the activity, “Normal Earthworm Responses to Threatening Environmental Stimuli – The Startle Response” (S#3)
- Discuss “Normal Earthworm Responses . . .” (S#3)
- Assign “Environmental Factors and Their Effect on Earthworm Movement” (S#4)

### Day 5

**Sensing and Responding/Student Reflections**

- Begin with “Environmental Change and Response to Change” (C#3) as an anticipatory exercise to “Sensing and Responding” (S#5). Read (S#5) in groups and discuss as a class. Refer to student work on normal earthworm crawling behavior and the “startle response”
- Assign “What Do You Think?” (S#6)

### Day 6

**Comparing Earthworm and Human Nervous Systems**

- Introduce the concept of a model
- Discuss how earthworms sense their environment and the function of the sensorimotor pathway, “Comparing Earthworm and Human Nervous Systems” (C#4)
- Discuss lowest observed effect concentration (LOEC)
- Discuss how chemicals in the soil can stimulate a behavioral response by an earthworm

### Day 7

**Experiment 1 – Simple Sensorimotor Reactions**

- Briefly review what students have learned about earthworm movement and the sensorimotor pathway
- Demonstrate to students how to make the test chamber
- Introduce *Experiment 1 – Simple Sensorimotor Reactions* to students and have them complete Experiment 1 Data Sheet (S#7)

### Day 8

**Experiment 2 – Preference Avoidance Behavior**

- Review what students learned in Experiment 1 from Day 7
- Introduce *Experiment 2 – Preference Avoidance Behavior* to students and have them complete Experiment 2 Data Sheet (S#8)

### Days 9-13

**Experiments 3&4 – Effect of Soil Type on Behavior Responses**

- Review what students learned in Experiment 2 from Day 8
- Introduce *Experiment 3&4 – Effect of Soil Type on Behavior Responses* to students and have them complete Experiment 3&4 Data Sheets (S#9, S#10)
- Complete “Observing Cation Exchange Capacity of Soils” (S#11)
- Assign “Sensorimotor Reactions and Behavior Responses” (S#12)
### Day 14

**VIDEO: EARTHWORM BURROWING AND NEURO-MUSCULAR INTERACTIONS**

- Discuss the possibility that environmental agents affect a worm’s locomotion and its survival
- Introduce and watch the *Introduction* and *Part I* of the video/DVD using the “Earthworm Video Guide (C#5) and relate to normal “Neuro-Muscular Interactions and Sensory Neurons” (C#2)
- Assign “Environmental Agents and Your Health” (S#13)

### Days 15-16

**VIDEO: OBSERVING THE EFFECT OF ALDICARB ON EARTHWORM BORROWING AND THE MEANING OFObserved EARTHWORM BEHAVIORS FOR HUMAN HEALTH**

- Finish and summarize observations from Day 12
- Do item “A” of “Pesky Pests, Pesky Thoughts?” (S#14)
- Read and discuss “A Is for Aldicarb, P Is for Pesticide” (S#15) and “How Does Aldicarb Work?” (S#16)
- Introduce and view the relevant segment of the video/DVD about burrowing behavior of aldicarb-treated earthworms
- Use “Burrowing Behavior of Aldicarb Exposed Earthworms” (S#17) to record qualitative and quantitative observations
- Use student questions to direct the class in an inquiry discussion of “What Might Happen If…?” (C#6)

### Optional

**STUDENT-DESIGNED EXPERIMENTS**

- Begin planning experiments on behaviors of earthworms treated with environmental chemicals using “Guidelines for Planning Your Experiment” (S#18)

*Note: An optional set of experiments, “Can Earthworms Learn?” (S#19), may be done and used as the basis for student-designed experiments related to earthworm learning in addition to those about earthworm locomotion. Although this option extends the time frame for the module 2 to 3 days, it increases student interest. Another option is to complete “What’s in a Word?” (S#20) extention activity.*

**IMPLEMENTATION OF STUDENT-DESIGNED EXPERIMENTS**

- Finish planning experiments
- Organize equipment and prepare solutions for student-planned experiments
- Carry out student-planned experiments and bring them to closure
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

ADVANCED PREPARATION GUIDE

MATERIALS PREPARATION

8 weeks (or more) before beginning the module
- Check inventory and order needed materials. (See Summary List of Materials, p. 14).
- Check with earthworm supplier on availability of earthworms – local bait shops are usually a good source of earthworms. Phone several to verify availability at the time you expect to do the module.
- Check with your school safety officer on using specific environmental chemicals. This is an important aspect of school safety and protects all those in the educational setting. Be sure to provide the safety officer with a list of possible environmental chemicals students are likely to suggest. This will prevent delays in implementing student investigations. Discuss with students the potential for environmental chemicals to affect organisms, including humans, and the safe use of them in the school setting. Then determine what substances are of interest to students.

2 weeks before beginning the module
- Arrange for purchase of earthworms – call your provider to confirm availability.
- Assemble needed materials. (See Summary List of Materials, p. 14)
- Have students bring in washed and rinsed plastic gallon jugs.
- Determine student groups – roles of members within a group will vary based on the activity.
### SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

**SUMMARY LIST OF MATERIALS FOR MODULE EXPERIMENTS**

**FOR THE CLASS:**

<table>
<thead>
<tr>
<th>Item</th>
<th>Quantity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dish soap, 1 bottle</td>
<td></td>
</tr>
<tr>
<td>Tape, 1 roll</td>
<td></td>
</tr>
<tr>
<td>Measuring Tape (metric)</td>
<td></td>
</tr>
<tr>
<td>Parafilm, 1 Roll</td>
<td></td>
</tr>
<tr>
<td>31 2-liter clean soda bottles</td>
<td></td>
</tr>
<tr>
<td>pH 4, 7 and 9 (use the strong acids H$_2$SO$_4$ or HNO$_3$ as models of acid rain—HCl works, too) Optional: Vinegar (a weak acid: use full strength and a 10% and 1% solution)</td>
<td></td>
</tr>
<tr>
<td>1000 parts per million (ppm) of: Copper, Manganese, Nickel, and/or Zinc</td>
<td></td>
</tr>
<tr>
<td>Distilled water</td>
<td></td>
</tr>
</tbody>
</table>

**FOR EACH GROUP:**

<table>
<thead>
<tr>
<th>Item</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Plastic gallon jug</td>
<td></td>
</tr>
<tr>
<td>Large bottle brush</td>
<td></td>
</tr>
<tr>
<td>Clean sheet of paper</td>
<td></td>
</tr>
<tr>
<td>Water</td>
<td></td>
</tr>
<tr>
<td>Clean cloth or sponge</td>
<td></td>
</tr>
<tr>
<td>Container to hold worms (12-16 oz plastic cup)</td>
<td></td>
</tr>
<tr>
<td>3 earthworms at room temperature</td>
<td></td>
</tr>
<tr>
<td>Moist paper towel</td>
<td></td>
</tr>
<tr>
<td>China marker or grease pencil</td>
<td></td>
</tr>
<tr>
<td>Dropper bottle of dechlorinated tap water</td>
<td></td>
</tr>
<tr>
<td>A clean, well-rinsed surface – a table, lab bench, or area of the floor</td>
<td></td>
</tr>
<tr>
<td>Dry, stiff paper toweling</td>
<td></td>
</tr>
<tr>
<td>Dissecting microscope with an illuminating light OR Magnifying glass with a strong flashlight</td>
<td></td>
</tr>
<tr>
<td>Long, skinny balloons</td>
<td></td>
</tr>
<tr>
<td>Compound microscope with objectives of 4X-40X</td>
<td></td>
</tr>
<tr>
<td>Glass rod</td>
<td></td>
</tr>
<tr>
<td>Matches</td>
<td></td>
</tr>
<tr>
<td>Small dropper bottle of 7% ethyl alcohol</td>
<td></td>
</tr>
<tr>
<td>Alcohol lamp or butane lighter for a cool flame</td>
<td></td>
</tr>
<tr>
<td>Narrow beamed halogen desk lamp, Maglight™, or microscope illuminator for intense beam of light</td>
<td></td>
</tr>
</tbody>
</table>
1 small ant farm

Potting soil and sand

1 container of red worms (approximately 36 worms/container)

1 plastic divider to divide soil into 2 sections

Test tube rack

6 test tubes (10 mL) per solution tested to create serial dilutions of 1000, 100, 10, 1, 0.1, 0.01 ppm + a separate tube with dechlorinated water (0 ppm)

Transfer pipets

Forceps

1 10 mL or 25 mL graduated cylinder

100 Pre-cut paper towels (5 cm x 5 cm)

1 squirt bottle and 1 liter beaker for rinsing worms between trials

Small dropper bottle of 0.2% methylene blue solution

Balance (measures to 0.1 g)

Weigh boats or paper

Spatula for weighing

Petri dishes

Ruler (in centimeters)

2 - 100 mL beakers

Red, yellow, and blue acetate transparencies cut into 2” squares to cover flashlights

Fine tip marking pen

2 stop watches

Ring stand

**FOR EACH STUDENT:**

- Apron
- Goggles
- Nitrile gloves (1 pair/student/class period)
## SUGGESTED LIST OF MATERIALS FOR STUDENT-DESIGNED EXPERIMENTS

<table>
<thead>
<tr>
<th>Item</th>
<th>Quantity</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 - 500 mL graduated cylinder</td>
<td>1</td>
</tr>
<tr>
<td>1 – 10 mL graduated cylinder</td>
<td>1</td>
</tr>
<tr>
<td>2 stopwatches</td>
<td></td>
</tr>
<tr>
<td>Moist potting soil</td>
<td></td>
</tr>
<tr>
<td>A deep container (test chamber) — 4” or more in depth</td>
<td></td>
</tr>
<tr>
<td>4 disposable containers (for exposure, e.g., plastic drinking cups)</td>
<td></td>
</tr>
<tr>
<td>4 worms</td>
<td></td>
</tr>
<tr>
<td>Aprons (1 per student)</td>
<td></td>
</tr>
<tr>
<td>Goggles (1 per student)</td>
<td></td>
</tr>
<tr>
<td>Nitrile gloves (1 pair per student)</td>
<td></td>
</tr>
<tr>
<td>Filter paper or paper towel (cut to fit bottom of plastic cups)</td>
<td></td>
</tr>
<tr>
<td>Dechlorinated tap water (1 gallon)</td>
<td></td>
</tr>
<tr>
<td>Balance</td>
<td></td>
</tr>
<tr>
<td>Spatula for weighing</td>
<td></td>
</tr>
<tr>
<td>Paper for weighing</td>
<td></td>
</tr>
<tr>
<td>Environmental agent</td>
<td></td>
</tr>
<tr>
<td>1 - 1 liter stock bottle with cap or flask with stopper, labeled</td>
<td></td>
</tr>
<tr>
<td>3 – 200 mL stock bottles with caps or flasks with stoppers, labeled</td>
<td></td>
</tr>
</tbody>
</table>
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

DAILY LESSON PLANS

INTRODUCTION

The Instructional Guide leads the teacher through the content and instructional aids integral to the module. Each class day is described using this sequence: Teacher Preparation For Class, Segue, Tasks To Be Completed During the Class Period, Materials, and Procedure. Detailed preparation and instructional guidelines for the teacher are provided here or in the cited part of the module. All class days are assumed to be 40-45 minutes in length. The module suggests few homework assignments.

Timing of the activities may be altered by the teacher. Please note the recommended instructional sequence progresses conceptually from normal locomotive behaviors to burrowing behavior as affected by various environmental agents. The guide ends with experiments designed by students. Their experiments are based on their questions about the effects of specific chemicals on earthworm locomotive behavior.
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

DAILY LESSON PLANS

DAILY LESSON PLANS PART I – CLASS DAYS 1 THROUGH 2:
A MODEL FOR NORMAL LOCOMOTION; UNDERSTANDING NORMAL EARTHWORM LOCOMOTION AND STRUCTURAL BASIS

This first set of lessons provides the background and the basic skills for the remaining sets of instructional days. Student background knowledge on earthworms is explored. Earthworm locomotion (function) and earthworm anatomy (structure) are related to neuro-muscular interactions. Instructional modes include both class and group discussions as well as laboratory activities involving data gathering and problem solving.

CLASS DAY 1 – INTRODUCING AND BEGINNING THE MODULE

Introduce the entire module and clearly state that its focus is on human environmental health. You may want to refer to an article in a local newspaper or magazine that tells about an environmental chemical and its effect on an organism. Ask students if they think the environmental chemical mentioned in the article might affect them, and if so, why and how.

Teacher Preparation for Class

1. Review “What Do I Know About Earthworms?” (Class Instructional Aid, C#1) and Student Activity Sheet “Earthworms!!!” (Student Activity, S#1).
2. Cut out the small spout of the plastic gallon jugs so the openings are 8-10 cm (3-4 inches) in diameter. This allows more surface area for chlorine to evaporate from the tap water during the 48 hour dechlorination period.
3. Assemble materials (see next page).

Tasks To Be Completed During the Class Period

- Direct student attention to the focus of the first part of the module, normal locomotion of earthworms and its structural basis, namely muscles and nerves.
- Use “What Do I Know About Earthworms?” (C#1) to elicit students’ prior knowledge about earthworms.
- Have student groups read “Earthworms!!!” (S#1) and discuss reasons for using dechlorinated water and keeping worms moist when experimenting with them. Check for understanding with brief class discussion.
- Wash plastic gallon jugs and prepare dechlorinated water while discussing the need for chlorine-free water.
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

DAILY LESSON PLANS

Materials

WORKSHEETS:
___ Activity Sheet “Earthworms!!” (S#1)

MATERIALS FOR THE CLASS:
___ Dish soap, 1 bottle
___ Tape, 1 roll

MATERIALS FOR EACH GROUP:
___ A pan containing moist soil and one earthworm
___ Plastic gallon jug
___ Large bottle brush
___ Clean sheet of paper

Procedure

1. Have one group of students wash the plastic gallon jugs with a diluted solution of dish soap, rinse them at least five times with tap water, and fill them 3/4 full with tap water.

2. Loosely tape cheese cloth over each gallon jug to keep out dust and allow the chlorine to volatilize out of the water.

3. Let filled gallon jugs stand for 48 hours to allow chlorine to volatilized from the water. Then put the cap back on the jug.

4. Keep dechlorinated water for future use (i.e., to moisten potting soil, keep the worms moist, and make solutions).
C#1 – “WHAT DO I KNOW ABOUT EARTHWORMS?” WEB ACTIVITY

Note to the Teacher: Ask students to share what they know about earthworms. Using a whiteboard, write words they mention and ask them to: a) link their word to others already shown, and b) explain the linkage. Or, have students come up and add their thoughts to the diagram. A web of terms connected by lines will result. After students have handled and observed worms at the beginning of S#1, the class may add to this web.
**S#1 – EARTHWORMS!!!**

*Note to the Teacher:* To stimulate student interest before they read about earthworms, give each student group a pan containing a live worm and moist soil. Have the group observe for a few minutes and then discuss their observations about worm movement, color, etc., and add to the web, C#1.

**READING**

“Nobody loves me. Everybody hates me. Guess I’ll go eat worms.” So the jingle goes. Even though the song is meant to humor us when we are feeling sorry for ourselves, it is important that we like both ourselves and earthworms. After all, earthworms are among our best friends!

Earthworms unknowingly help us as they burrow underground to create their homes. In creating their tunneled homes, they mix and enrich the soil and make passages for air, water, and other organisms. Scientists estimate that there are about 50,000 earthworms in each acre of moist soil, or slightly more than one earthworm for about every square foot of ground. A soil with lots of dead plant and animal matter may contain over 20 earthworms per square foot. Yet how many of us think about them and all the good things they do every time we take a step outdoors?

We probably do not think of them because we normally do not see them when we are outdoors. However, if we look carefully, we may see earthworms on the ground at night when temperatures are moderate, light level is very low and the air is moist. Otherwise, they hibernate underground to survive cold or dry weather, or move through the soil to eat. As they eat they create tunnels. To make tunnels, the worm’s muscular prostomium (first segment) wedges into the soil and eats it. The muscular pharynx (throat) pushes the soil and any mixed-in dead plant matter further into the worm’s digestive tract. When the food and soil pass through the organs of the digestive tract, they are mixed and broken down into nutrients. The nutrient mix becomes available to plant roots once the worm excretes its wastes as “castings.” If you look carefully, you may find some castings, small coiled knots, on the ground near the opening of a worm burrow. Whether above or below ground, castings contribute to soil structure and fertility, providing the conditions and nutrients so important for plant growth.

Important to plants and other organisms are the air and water that enter the soil through earthworm burrows. Air is important for maintaining healthy plant roots and beneficial soil bacteria. Air in the soil is also very important to earthworms because they breathe through their moist skins. If the soil becomes water-logged, earthworms must move out of their burrows onto the ground to avoid drowning. Yet soil must contain some water if the earthworms’ skins are to be kept moist for breathing and moving (locomotion).

An earthworm moves through soil or on the ground using two sets of muscles: long ones that run the length of its body and ring-shaped ones that circle each body segment. When the long
S#1 – EARTHWORMS!!!

muscles of the worm contract, the body of the earthworm becomes shorter and thicker. When the worm’s ring-shaped muscles contract, its body becomes longer and thinner. When you watch an earthworm crawl, you will notice that parts of the body may be thick and other parts are thin. You may also notice that the thickness of a given section of the body changes from thick to thin, and back to thick! For example, when the worm moves forward, its front extends forward and gets thin. Then, as the rear of the body is pulled forward, the head end thickens and the portion behind gets thinner. As the thinning and thickening pass along the length of its body, you can observe the wave-like pattern so characteristic of earthworm locomotion.

Because earthworms do not move great distances, a person wanting to improve soil needs to provide better conditions for earthworm health and reproduction. One effective way to improve soil is to increase the amount of dead plant material available to worms. Dead leaves or plant material may be layered on the soil as mulch or worked into the soil. Mulch will help hold soil moisture and temperature at levels favorable to earthworm activity. Working plant material into the soil provides food for worms, although it temporarily disturbs them. Another way to improve conditions for earthworms is to keep soil at a neutral pH. As the earthworm population increases, it will help maintain this desirable pH. The worms improve not only their environment, but ours as well! As long as they are fed, natural worm populations increase.

Earthworm populations are maintained or increased by a most interesting form of reproduction. While each earthworm has both male and female reproductive organs, most earthworms are not self-fertile; that is, the eggs of one earthworm cannot be fertilized by the sperm of that same earthworm. To reproduce, earthworms must mate. When earthworms are ready to mate, the largest body segment, the clitellum, changes from pinkish to red-orange in color. Two earthworms meet head to head and move along one another so their clitella are somewhat separated. Mucus is then secreted from the clitellum of each worm and the sperm from each worm moves into the storage pouch of the other worm. The mucus dries and forms a capsule into which more fluid is secreted. The worm then backs out of its capsule. As it does so, first the eggs and then sperm are deposited into the capsule; finally fertilization occurs. When the worm has fully left the capsule, the capsule remains in the soil and the fertilized eggs develop into baby worms.

Several factors are known to affect earthworm populations. Those already mentioned are soil temperature and moisture, soil pH, and the amount of dead plant matter available for food. Research has demonstrated that plowed fields have fewer earthworms than do pastures. Scientists have shown that placing fertilizer and insecticides in the soil when planting a row crop decreases worm populations near the row. As might be expected, the effect of agricultural chemicals on earthworm populations is different for each chemical. Chemicals like the pesticide aldicarb are known to be highly toxic to earthworms. As you progress through the module, you will observe the effect of aldicarb on earthworms. Earthworms respond to the chemicals that are dissolved in the water that surrounds soil particles. They may be crawl towards or away from these chemicals. While some of these chemicals, such as copper, manganese or zinc, are important in small amounts, they become toxic at higher concentrations. In this module, you will also observe how earthworms respond to different concentrations of these metal salts, as well as changes in soil pH (a measure of acidity).
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

CLASS DAY 2 – EXAMINING EARTHWORM ANATOMY AND NEURO-MUSCULAR INTERACTIONS

Teacher Preparation for Class

1. Review “Earthworm Structures and the Work They Do” (S#2A/2B).
2. Get worms out of refrigerator, place moist paper towel in container, put two worms on paper towel and gently cover with moist paper towel. Set up as many containers as you have groups.
3. Set out materials for group work on external and internal anatomy.

Procedure
See Activity Sheet “Earthworm Structures and the Work They Do” (S#2A/2B).

Segue
Have students share what they know about how an earthworm moves.

Tasks To Be Completed During the Class Period
- Begin lab, “Earthworm Structures and the Work They Do” (S#2A/#2B).

Materials

WORKSHEET:

“Earthworm Structures and the Work They Do” (S#2A/#2B)

FOR EACH GROUP:

FOR EXTERNAL ANATOMY

- A clean, well-rinsed surface – a table, lab bench, or area of the floor
- 1 or 2 live earthworms in moist paper toweling in container
- Dropper bottle of dechlorinated water
- Water
- Clean cloth or sponge
- Dry, stiff paper towel
- Activity sheet “Earthworm Structures and the Work They Do” (S#2A/#2B)
- Dissecting microscope with an illuminating light OR magnifying glass with a strong flashlight
FOR INTERNAL ANATOMY

- Long, skinny balloons
- Compound microscope with objectives of 4X-40X
- Prepared microscope slide of earthworm cross sections

Procedure

See Procedure Activity Sheet “Earthworm Structures and The Work They Do” (S#2A/S#2B).
S#2A/2B – EARTHWORM STRUCTURES AND THE WORK THEY DO

Even though the earthworm looks simple, it has a fairly complex set of body systems and is able to appropriately respond to many different stimuli. In this activity, we are going to see how its body structures enable it to move. We are going to examine worm anatomy and see that underlying the worm’s simple appearance are some surprisingly complex structures - structures extremely well-adapted to the animal’s soil environment. First you will examine the external structure of the worm and relate observed structures to the worm’s crawling behavior. Then you will study its internal structures and relate those to its crawling behavior. Finally, you will summarize what you have learned about earthworm structures and their importance to the earthworm’s crawling motion and survival.

Before beginning the activity, look at the grid below. Enter information you already know. As you do the activity and readings, add information to complete the grid.

GRID: COMPARING ANIMAL MOVEMENT AND SUPPORT

<table>
<thead>
<tr>
<th></th>
<th>MOVEMENT</th>
<th>SUPPORT</th>
</tr>
</thead>
<tbody>
<tr>
<td>EARTHWORM</td>
<td>Moves by contracting:</td>
<td>Is supported by:</td>
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<td></td>
<td>and uses: ________________ to grip soil.</td>
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<tr>
<td>CATERPILLAR</td>
<td>Moves by contracting:</td>
<td>Is supported by:</td>
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<td>________________________________________________________________________</td>
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<td></td>
<td>and uses: ________________ to grip soil.</td>
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<tr>
<td>SNAKE</td>
<td>Moves by contracting:</td>
<td>Is supported by:</td>
</tr>
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<td></td>
<td>and uses: ________________ to grip soil.</td>
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<tr>
<td>HUMAN</td>
<td>Moves by contracting:</td>
<td>Is supported by:</td>
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</tbody>
</table>
S#2A/2B – EARTHWORM STRUCTURES AND THE WORK THEY DO

DIRECTIONS:

• Before beginning the activity, clean and rinse your work area well.
• Have all materials available.
• Group members should take turns reading directions aloud.
• ALL group members will do each step of the procedure and record.

REMINDEERS:

• Do not touch the worms more than necessary.
• Keep your worms damp so they do not dry out and suffocate.

MATERIALS (FOR EACH STUDENT GROUP)

For External Anatomy

- Water
- Clean cloth or sponge
- Activity sheet
- Dry, stiff paper towel
- 1 or 2 live earthworms in moist paper toweling in a paper cup
- Dropper bottle of dechlorinated tap water to moisten worms
- Dissecting microscope with an illuminating light
- Magnifying glass with a strong flashlight

For Internal Anatomy

- Long, skinny balloons
- Compound microscope with objectives of 4X - 40X
- Prepared microscope slides of earthworm cross sections
1. As you work through this procedure, label the diagram at the end of this section with the underlined names provided in the directions below.

2. Clean your work area well with fresh water and a clean cloth or sponge. Put the live earthworm on your work area surface and carefully examine it with the magnifying glass. The most obvious characteristic of the worm is that its body is made up of a series of similar looking ringed structures. Each ringed structure is called a segment and it is this characteristic that gives these worms their name. (We are also made of repeating segmented structures. In our case, our body segments are different, but you can still see them in the repeating bones of our spines and ribs. Segmentation is a common feature of the body plan of many animal groups.) In the worm there is one segment that is much longer and thicker than the others. It is called the clitellum and is involved in sexual reproduction of earthworms. Worms are hermaphrodites (they are both male and female), yet they generally mate with another worm.

3. Carefully watch the earthworm move; you can probably tell the color and shape differences between the head and tail ends of the worm. Carefully examine the last segment and first segment (prostomium) of the worm. What are the differences?

What are the jobs of the head end of the worm? How might those jobs account for the shape differences you noted above?

4. Touch and look at the top (dorsal) of the worm, and the bottom (ventral) of the worm. Note three differences.

A.

B.

C.

It is important for you to note these differences because you will use them later as you observe earthworm behavior.
5. Use your fingers and gently flip the worm upside down. What does it do?

Is its motion smooth or jerky?

Keep a picture of this motion in mind. You will use it in later activities. This shows us another behavior of the worm. It can flip itself over onto its underside/front. (Interestingly, this same type of behavior is a developmental milestone of human babies. They first develop enough strength to flip themselves over onto their backs, but it takes them longer to be able to flip from their backs onto their front sides.) The ability of the worm to perform this behavior shows us that at least the worm can tell its back from its front.

6. Put the worm on a stiff dry paper towel. When it starts moving, put your ear close to the worm and listen. What do you hear?

The earthworm maintains traction with the ground by means of many fine bristles (the *setae*) on its underside/front. Rub the worm’s underside with your finger from head to tail. Then rub from tail to head? Note any differences.

Does the roughness change when the worm changes shape? What might control this?

7. Let the worm crawl and watch the setae as they and the earthworm move. Use the magnifying glass and a bright light to observe the setae. How are they shaped?

How does the movement of these fine bristles help the worm in its environment?

In addition to providing supports, muscles work to extend the setae from the worm’s body and to pull them back again. How many setae are in each segment? To answer this, you may need to observe the setae with the dissecting microscope.

Draw and label a few *setae* on the diagram of the worm.
8. Place the worm in moist paper toweling in the container and return it to the appropriate place.

9. Enter the information you have learned so far in the grid at the beginning of this exercise and be sure you label the worm below.

**FIGURE 1: EARTHWORM EXTERNAL ANATOMY**
BACKGROUND

In studying the locomotion of the worm, it was evident that the worm moved in waves of muscular contractions that passed smoothly along its body. You observed waves of alternate lengthening and shortening of a portion of the worm’s body. The lengthening is caused by contraction of circular muscles that squeeze on the inside of the worm to make it thinner and longer. The shortening and thickening are caused by longitudinal muscles that pull the extended portion of the worm together. Before looking at those muscles under the microscope, think about how they cause movement. All muscles must be attached to a structure to cause movement. Human muscles work on bones to which they are attached; or in the case of intestines, the muscles work on the food in the gut at the same time they are held in place by connective tissues.

From handling worms, you know there are no bones in a worm. What do the worm’s muscles attach to and work on? They attach to and work on a water skeleton. The water skeleton is found between the muscles and the gut and runs the entire length of the animal. Label the water skeleton in Figure 2. Then follow the procedure below to understand how the muscles work as the worm moves.

1. Take a long balloon, fill it with water, and tie a knot to close off the balloon. Put your hands around the outside of the balloon and squeeze it. This is the same action that the circular muscles have on the fluid-filled “skeleton” of the worm. What two things happen to the balloon as you squeeze?

   A. 

   B. 

   What would happen to the body of the worm?

If you let go of the balloon, what does it do?

The worm’s body does not behave in this way. Instead, it must use its longitudinal muscles to return to its original shape. This flexible, water-filled “skeleton” of the worm actually makes it possible for the worm to crawl through the soil. Using the water skeleton to push against, the worm can move its head into small openings. It makes those openings larger by thickening its body. It then pulls forward into the enlarged opening. This is the worm’s perfect solution for moving through its environment.
2. Put the slide of the earthworm cross-section on the microscope stage and make sure the light is shining through the thin slice of worm. Begin examining the cross-section with the lowest power objective (usually 4X). You can probably see the entire cross-section of the worm. The first thing to notice is that the top and bottom of the worm are different. The **dorsal blood vessel** or heart is on the back (top) of the animal and the **nerve cord** is on its ventral (bottom). Around the outside of the animal are two layers of muscles: the **outer circular muscles and the inner longitudinal muscles**. We know they do different jobs to move the earthworm. Describe any differences in their structure. Relate the process of squeezing the water balloon to each of these muscle layers.

3. Locate the **nerve cord** of the animal. It is fairly small and located on the opposite side of the animal from the heart or dorsal blood vessel. Once you have located it with a low power objective, use higher power objectives to examine it. The nerve cord is the communication system of the animal. It triggers the muscles to act and coordinates action among neighboring segments. The nerve cord also receives and analyzes sensory signals so the animal can respond appropriately.

   Why do you think the nerve cord is on the ventral (bottom) side? Compare and contrast to where the nerve cord is in snakes.

4. Develop a testable hypothesis to explain any differences.
The most obvious structures in the worm’s nerve cord are the three large circular shapes near its top. These are extremely large nerve fibers called giant axons. Most nerve fibers are very small. The giant axons’ large size allows them to send electrical signals extremely rapidly – much more quickly than the smaller nerve fibers. In fact, they are the nerve fibers involved in the escape or “startle response” so very important for the worm’s survival. (Try to estimate the size of the giant axons. You can do this by measuring the width of the entire worm slice with a ruler directly from the slide and estimating how large the nerve cord might be and then the giant axons themselves.

5. Later in the module, you may choose to investigate whether an environmental chemical affects the life-saving “startle response” of the worm. If it does, either the giant axons or the muscles themselves will have been affected by the chemical.

6. Because two earthworms need to be lined up in a particular fashion to reproduce, what would you expect to happen to the earthworm population if a chemical affected their ability to line up?
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

DAILY LESSON PLANS PART II – CLASS DAYS 3 THROUGH 11:
OBSERVING NORMAL EARTHWORM RESPONSES TO THREATENING STIMULI,
COMPARING EARTHWORM AND HUMAN NERVOUS SYSTEMS, UNDERSTANDING THE
EFFECT OF ENVIRONMENTAL AGENTS ON BURROWING, AND ARTICULATING
POSSIBLE IMPLICATIONS FOR HUMAN HEALTH

CLASS DAYS 3-4: EXAMINING NERVE-TO-MUSCLE SIGNALING AND OBSERVATION
OF NORMAL EARTHWORM RESPONSES TO THREATENING ENVIRONMENTAL
STIMULI

Teacher Preparation for Class

1. Set up materials needed to finish “Earthworm Structures and the Work They Do” (S#2A/#2B) if not completed in previous day.
2. Review “Nerve-Muscle Interactions and Sensory Neurons” (C#2).
3. Review “Normal Earthworm Responses to Threatening Environmental Stimuli – The Startle Response” (S#3) and “Environmental Factors and Their Effect on Earthworm Movement” (S#4).
4. Get worms out of refrigerator, place moist paper towel in container, put three worms on paper towel, and gently cover with paper towel. Set up as many containers as you have groups. Set up remaining required materials.
5. Be sure to submit the list of student-suggested chemicals to the school safety officer for evaluation.

Procedure

Introduce and discuss “Nerve-Muscle Interactions and Sensory Neurons” (C#2). Use concepts from this discussion to introduce, “Normal Earthworm Responses to Threatening Environmental Stimuli – the Startle Response” (S#3).

Segue

Briefly review external anatomy of the earthworm and any internal anatomy covered, and relate these to earthworm movements. Prompt students to share what they know about earthworm movements, explain the neuro-muscular basis of earthworm movement (C#2), discuss that one must know what normal movements (control) look like in order to determine whether chemicals interfere with normal movements. During this discussion, review concepts previously introduced. Clarify that the day’s activity establishes an understanding of the earthworms’ normal escape or “startle response.”

Tasks To Be Completed During the Two Class Periods

- Finish and discuss “Earthworm Structures and the Work They Do” (S#2A/S#2B).
- Introduce and discuss normal “Nerve-Muscle Interactions and Sensory Neurons” (C#2).
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

- Explain and complete the activity, “Normal Earthworm Responses to Threatening Environmental Stimuli – The Startle Response” (S#3).
- Assign “Environmental Factors and Their Effect on Earthworm Movement” (S#4).

NOTE: To save time, the set of tests in this procedure may be divided among groups in the class so each group does A and B or A and C instead of the entire set of three tests.

EXPERIMENT

MATERIALS

WORKSHEETS:

- “Nerve-Muscle Interactions and Sensory Neurons” (C#2)
- “Normal Earthworm Responses to Threatening Stimuli – The Startle Response” (S#3)
- “Environmental Factors and Their Effect on Earthworm Movement” – Assignment (S#4)

FOR EACH GROUP:

- Container (paper cup) with 3 earthworms in moist paper toweling at room temperature
- Dropper bottle of dechlorinated water
- Water
- Clean cloth or sponge
- Glass rod
- Matches
- Small dropper bottle of 7% ethyl alcohol
- Alcohol lamp or butane lighter for a cool flame
- Narrow beamed halogen desk lamp, Maglight™, or microscope illuminator for intense beam of light
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

C#2 – NERVE-MUSCLE INTERACTIONS AND SENSORY NEURONS

ILLUSTRATION OF NERVE-TO-MUSCLE SIGNALING

To the right is an electron microscope photograph of a motor neuron connecting to a muscle, the region called the **neuromuscular junction**. The vesicles will release a chemical messenger (acetylcholine) into the space or synaptic cleft between the nerve ending and the muscle fibers. The chemical messenger causes the muscles to contract and allow the animal to move. Below is a diagram to help you understand what this photograph is showing.

Source: Kimball’s Biology Pages (http://biology-pages.info)

Source: http://anatomycorner.com/muscles/notes_muscles.html
This diagram shows a nerve cell that signals a muscle at the synapse or neuromuscular junction.

This diagram illustrates the normal sequence of events at a nerve-muscle synapse.

a) Chemical messenger (green half-capsules) is released at the end of the nerve axon (light brown) into the synapse (gap) between the nerve ending and the receiving, relaxed muscle (dark pink showing less overlap between filaments) with receptor sites (yellow).

b) Chemical messenger binds to the receptor sites and stimulates the muscle to contract (overlap between filaments increases and muscle contracts).

c) The enzyme associated with the synapse (light blue arcs on the muscle) breaks down the chemical messenger and the muscle relaxes (overlap between filaments returns to original state).
S#3 – NORMAL EARTHWORM RESPONSES TO THREATENING STIMULI – THE “STARTLE RESPONSE”

As the earthworm crawls along, it responds to its environment. Like us, it depends on both its environment and its ability to respond to stimuli in its environment. Some things in a worm’s environment that are important to it are not necessarily important to us and vice versa. What is important to understand is that we and earthworms are interdependent, so what affects the worm will to some extent affect us.

In later activities you will learn about specific environmental chemicals and their effect on the earthworm’s ability to respond. This activity allows you to observe normal earthworm responses to several stimuli. Later, when you check out the possible effect of a chemical, you will already know what normal behavior looks like. You will then be able to compare behaviors of untreated and treated worms. Remember, the earthworm’s nerve-muscle interactions are similar to yours, so the earthworm is a model for understanding possible human health effects.

Below is a Prediction / Reflection Log. First read the questions in the “Tests” column. Then think about each test and write your expectation of how the earthworm will respond to that test in the “My Hypothesis” column. When you have finished each test, return to the log and complete the column, “My Observations of Earthworm Behavior.”

**Prediction / Reflection Log**

<table>
<thead>
<tr>
<th>WHAT DO I ALREADY KNOW?</th>
<th>WHAT DO I NEED TO KNOW (HYPOTHESIS DEVELOPMENT)?</th>
<th>WHAT DO I DO TO LEARN WHAT I NEED TO KNOW (METHODS DEVELOPMENT)?</th>
<th>WHAT DO I NOW KNOW (OBSERVATIONS AND DATA COLLECTION)?</th>
<th>WHAT DO I NOW KNOW (DATA ANALYSIS AND INTERPRETATION)?</th>
<th>WHAT DO I NOW NEED TO KNOW (NEXT EXPERIMENTS)?</th>
</tr>
</thead>
<tbody>
<tr>
<td>WHERE DO WORMS LIVE?</td>
<td></td>
<td>What will an earthworm do when light is shone in its path? Behind it? On top of it? Different color light?</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HOW DO WORMS KNOW WHAT IS BAD FOR THEM?</td>
<td></td>
<td>What will an earthworm do when alcohol is placed on/near its head?</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HOW DO WORMS KNOW IT IS HOT OUTSIDE?</td>
<td></td>
<td>How will an earthworm respond when a heated object is placed by its head/tail?</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Before beginning, clean and rinse your work area well. Have all materials available and choose roles: Worm Handler, Equipment Manager, and Recorder. All will keep records. Switch roles at the end of each section of the procedure.

REMINdERS:

- Do not touch the worms too much because they will stop responding. This is the simple kind of learning known as habituation.
- Once you start this set of experiments, use one worm until it stops responding and then switch to another worm while the first one recovers.
- Keep your worms damp or they will dry out and suffocate.

MATERIALS:

- Container (paper cup) with three earthworms in moist paper toweling at room temperature
- Water
- Clean cloth or sponge
- Glass rod
- Dropper of dechlorinated tap water to keep animals moist
- Matches
- Small dropper bottle of 7% ethanol (ethyl alcohol)
- Narrow-beamed halogen desk lamp, Maglight™ or microscope illuminator for intense beam of light
- Alcohol lamp or a butane lighter for a cool flame
A. PROCEDURE: LIGHT STIMULUS

1. Read questions 1 through 6 before you proceed.
2. **Equipment Manager:** Clean and rinse your work area well.
3. **Worm Handler:** Put a worm on your work area and let it crawl. Make sure it is moist.
4. **Equipment Manager:** When the worm is moving along nicely, shine a narrow flashlight beam in the path ahead of the worm, *not on the worm.*
5. **All:** Carefully observe what the worm does when its head enters the beam of light and discuss the worm’s response as a group.
6. **Recorder:** Summarize the group’s observation, and complete item 1 in the “Observations” section. Let the worm rest for a minute.
7. **Equipment Manager:** Shine the beam *on the worm’s head.*
8. **All:** Carefully observe the worm’s response to the light shone on its head and discuss your observations.
9. **Recorder:** Summarize the group’s observation, and do item 2.
10. As a group, complete all items.

**OBSERVATIONS: RESPONSE TO LIGHT STIMULUS**

1. How does the earthworm respond when it’s head enters the light?

   ____________________________________________

2. How does the earthworm respond when the light beam is directed at the middle of its head?

   ____________________________________________

3. Think about where the worm lives and what it does to survive. Does its response to light make sense? Explain.

   ____________________________________________
4. How is this response important for the worm’s survival?

5. If the worm had vision like ours, would that be helpful for its survival?

6. Can you think of reasons why vision like ours might actually be harmful to the worm’s chances to survive in its environment?

B. PROCEDURE: CHEMICAL STIMULUS

1. Read questions 1 through 3 before you proceed.

2. **Worm handler:** Put a worm on your work area and let it crawl. Make sure it is moist.

3. **Equipment manager:** When the worm is moving along nicely, put a single drop of 7% **ethanol** on the worm’s head.

4. **All:** Carefully observe what the worm does when the alcohol is put on its head and discuss what you observed.

5. Record what you observe and complete questions 1 - 3 in “Observations.”

**OBSERVATIONS: RESPONSE TO ALCOHOL/CHEMICAL STIMULUS**

1. How does the worm respond to the drop of alcohol placed on its head?

2. How is this behavior similar to its response to light shone on its head?
S#3 – NORMAL EARTHWORM RESPONSES TO THREATENING STIMULI

3. How is this behavior different from that to light shone on its head?

C. PROCEDURE: HEAT STIMULUS

1. Read questions 1 through 5 before you proceed.
2. Equipment Manager: Clean and rinse your work area well.
3. Equipment Manager: Light the alcohol lamp or butane lighter and warm the glass rod in the flame. Do not melt the glass.
4. Worm Handler: Put a worm on your work area and let it crawl. Make sure it is moist.
5. Equipment Manager: When the worm is moving along nicely, bring the rod close to the head of the worm but do not touch the worm’s skin.
6. All: Observe the worm’s response as a group. Discuss what you observe. Then record your discussion. Complete items in the “Observations” sections below.

OBSERVATIONS: RESPONSE TO HEAT

1. How does the worm respond to the warm rod placed near its head?
2. How is this behavior like the worm’s response to light shone on its head?
3. How is this behavior different from its response to light shone on its head?
4. How is this behavior like the worm’s response to ethanol placed on its head?
5. How is this behavior different from its response to ethanol?

GENERAL OBSERVATIONS

1. How are these responses different from crawling?

2. Complete the second column of the Prediction / Reflection Log found at the beginning of this activity. Then answer questions 1 – 3 below:

HOW WOULD YOU RESPOND TO THESE STIMULI?

1. Do you find some of these stimuli irritating or harmful?

2. Describe how you would respond to one of these stimuli.

3. Review your results from this activity and tell if you have changed your mind about whether some environmental factors affect earthworm movement.

REMEMBER:
COMPLETE THE LOG AT THE BEGINNING OF THIS ACTIVITY and write a general description of the earthworm’s response to the three stimuli below or on the back of this page.
**DIRECTIONS**

Look at the information in the grid below. As you think about each factor, place a check in either the “yes,” “no” or “uncertain” column to indicate your expectation of whether each factor would affect the worm’s speed.

**ENVIRONMENTAL FACTORS AND THEIR EFFECT ON EARTHWORM MOVEMENT**

<table>
<thead>
<tr>
<th>FACTOR</th>
<th>WOULD THIS AFFECT WORM MOVEMENT?</th>
</tr>
</thead>
<tbody>
<tr>
<td>Would the movement of an earthworm be affected by…</td>
<td>YES</td>
</tr>
<tr>
<td>The temperature of the soil and air around the worm?</td>
<td></td>
</tr>
<tr>
<td>The amount of light in the worm’s environment?</td>
<td></td>
</tr>
<tr>
<td>The amount of moisture on the ground and in the soil?</td>
<td></td>
</tr>
<tr>
<td>Chemicals that might be in the soil and the worm’s food?</td>
<td></td>
</tr>
<tr>
<td>The location of the earthworm in the soil or the ground?</td>
<td></td>
</tr>
</tbody>
</table>

**WITH A BRIEF STATEMENT, EXPLAIN TWO OF YOUR CHOICES.**

1. _______________________________________________________________________

__________________________________________________________________________

2. _______________________________________________________________________

__________________________________________________________________________
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

CLASS DAY 5 – SENSING AND RESPONDING/STUDENT REFLECTIONS

Teacher Preparation for Class

1. Set out materials needed to finish the activity sheet “Normal Earthworm Responses to Threatening Stimuli – The Startle Response” (S#3) if necessary.

2. Review “Environmental Change and Response to Change” (C#3), “Sensing and Responding” (S#5), and “What Do You Think?” (S#6). Relate the structures and their work to the worms’ survival and the possible effect of environmental agents on earthworm locomotion and survival.

Segue

Discuss observations to date on “Normal Earthworm Responses to Threatening Stimuli – The Startle Response” (S#3).

Tasks To Be Completed During the Class Period

- If necessary, finish “Normal Earthworm Responses…” (S#3), discuss, and summarize. Compare the “startle response” to normal crawling behavior.
- Begin with “Environmental Change and Response to Change” (C#3) as an anticipatory exercise to the reading “Sensing and Responding” (S#5). Read and discuss in groups and then as a class.
- Introduce the assignment “What Do You Think?” (S#6). Students may refer to the Prediction/Reflection Log in Activity Sheet (S#3).
- If time permits, segue to possible effects of environmental chemicals on burrowing behavior.

Assignment

- Complete “What Do You Think?” (S#6).

MATERIALS

WORKSHEETS:

- “Environmental Change and Response To It” (C#3)
- “Sensing and Responding” (S#5)
- “What Do You Think?” (S#6)
Note to the Teacher: This class activity may be used as an anticipatory exercise for the reading “Sensing and Responding.” Its content will heighten student awareness of an organism’s ability or inability to respond to an environmental change, regardless of whether it is potentially beneficial or harmful to the organism.

If the class cannot complete an item, leave it blank and revisit the activity after the reading. Be sure to discuss the responsibility of individuals and governments to be informed about possible and actual environmental change.

**DIRECTIONS:**
The first chart lists some changes in the human environment.

1. Complete the items in the second column by indicating whether the change would be detected by any of our senses.
2. In the third column briefly describe how a person might respond to each environmental change.

<table>
<thead>
<tr>
<th>ENVIRONMENTAL CHANGE</th>
<th>DETECTABLE</th>
<th>RESPONSE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Popcorn vapor in air</td>
<td>Yes</td>
<td>Go buy popcorn</td>
</tr>
<tr>
<td>Weedkiller Atrazine</td>
<td>No</td>
<td>Carefully use it</td>
</tr>
<tr>
<td>Chocolate vapor in air</td>
<td>Yes</td>
<td>Buy and eat a chocolate bar</td>
</tr>
<tr>
<td>Toe stubbed</td>
<td>Yes</td>
<td>Yell “ouch”</td>
</tr>
<tr>
<td>CO in air</td>
<td>No</td>
<td>N/Pass out</td>
</tr>
<tr>
<td>Screech of car brakes</td>
<td>Yes</td>
<td>Turn toward the noise</td>
</tr>
<tr>
<td>Increased UV rays from sun</td>
<td>No</td>
<td>Get sun burn or skin cancer</td>
</tr>
</tbody>
</table>
The second chart lists some changes that might occur in an earthworm’s environment. Complete this chart in the same way you completed the first chart.

<table>
<thead>
<tr>
<th>ENVIRONMENTAL CHANGE</th>
<th>DETECTABLE</th>
<th>RESPONSE</th>
</tr>
</thead>
<tbody>
<tr>
<td>(Yes or No)</td>
<td>(Describe it)</td>
<td></td>
</tr>
<tr>
<td>Touch to head</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Heat near body</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Light in path</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Aldicarb in soil</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
S5 – SENSING AND RESPONDING

Anticipation Questions:

1. What signals do you pick up from (stimuli do you detect in) your environment?
2. Which of these signals (stimuli) is beneficial? Harmful? How do you respond to them?
3. Is it likely you are unaware of some signals from or changes in your environment?
4. Are you able to respond to stimuli/changes you do not know about? How do you feel about that?

As the earthworm crawls along, it responds to its environment. The worm’s survival, like ours, depends on its ability to respond to what happens in its environment. While worms and humans are able to respond to many changes in the environment, neither is able to respond to all changes. Naturally, human and earthworm responses are limited to the stimuli each is able to sense. Some stimuli in a worm’s environment that are important to it are not necessarily important to us and vice versa. For example, an earthworm has special cells in its skin that detect vibrations in the soil made by its predators. Humans cannot detect these vibrations. Generally, each life form has a range or set of stimuli that it can naturally sense and respond to.

Usually stimuli that an organism can sense are important for its survival in a natural setting. These detectable stimuli may be threatening or non-threatening. The proper responses to them usually contribute to the survival of the organism. Harmful stimuli are usually avoided. But if an organism cannot sense a harmful change in the environment, it cannot respond. For that organism, it is as if there has been no change in the environment – unless the life form can use its intellect/intelligence to understand that a change has taken place. Only with knowledge can an organism respond to environmental changes it is unable to sense. If the change is beneficial to the organism, survival is not an issue. But if the change is potentially harmful, only the informed organism understands and has the opportunity to act to eliminate the undetectable harm or avoid its dangers. Unlike most other organisms, we humans are capable of informing ourselves. How do we deal with substances we cannot sense but know to be harmful?

Carbon monoxide serves as an example. It is a harmful substance we cannot sense; we are unable to see, smell or taste this gas, yet it can kill us because it interferes with delivery of oxygen to our cells. For centuries coal miners in particular have been aware they could die from working in underground mines. Without knowing what in particular was causing their symptoms of achy muscles, fatigue, headache, dizziness, disorientation, and even death, miners learned to use a more sensitive animal to warn them of possible harm. Specifically, they carried canaries or mice...
that were highly-sensitive to carbon monoxide into the coal mines to warn of danger. If the canary or mouse fainted, miners knew they were in great danger and needed to leave the mine.

Today, mines and other workplaces are outfitted with devices that detect carbon monoxide. In our homes we protect ourselves by properly installing and maintaining appliances that burn natural gas, oil or kerosene. Our public health and fire departments help protect us by telling us that if all the members of a family are headachy, fatigued, dizzy or disoriented, carbon monoxide poisoning might be the cause and the family should seek immediate medical help. By knowing the symptoms of carbon monoxide poisoning and seeking help when we experience them, we are able to protect ourselves from a harmful substance we cannot sense. Humans are fortunate to have the brainpower to protect themselves from harm they cannot detect with their senses. But most organisms have little ability to respond and protect themselves if they cannot sense a harmful change in their environment.

In the investigation that follows this reading, you will observe the worm’s response to harmful stimuli it senses. In later activities you will learn about specific environmental chemicals and their effect on the earthworm’s ability to respond. As you do these activities, think about the earthworm’s ability to protect itself from the chemicals to which it is exposed. Also think about your ability to detect and protect yourself from the chemicals to which you are exposed. Are you aware of what you are exposed to? If you are aware, is it because your senses detected and informed you or because of independent information provided to you? This is why warning labels and symbols like “Mr. Yuck” and the skull and crossbones are on products. What do you do about these exposures? Remember, the earthworm’s nerve-muscle interactions are similar to yours. What affects their nerve-muscle interactions may affect yours. The earthworm is a model for understanding the possible human health effects from exposure to environmental substances.
1. Now that you have tested the earthworm’s response to three different threatening stimuli, think about how important it is for the worm to be able to both sense and respond to these stimuli. Using what you have learned, write a short paragraph that answers the following questions: Why is it important for the earthworm to be able to sense and respond to stimuli? What would it mean for the earthworm if it were unable to detect or respond to a threatening stimulus?

2. Do you think there are stimuli in the earthworm’s environment that it is unable to detect or respond to? Explain.

3. Are there things in the earthworm’s environment that affect its ability to sense or respond to stimuli? Explain.

4. If there are substances in its environment that affect either the worm’s ability to sense or respond to a stimulus, what might that mean for the earthworm?

5. If there are substances in our environment that affect our ability to sense or respond to a stimulus, what might that mean for us?
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

CLASS DAY 6 – COMPARING EARTHWORM AND HUMAN NERVOUS SYSTEMS

Teacher Preparation for Class

1. Review “Environmental Change and Response to Change” (C#3), “Sensing and Responding” (S#5), and “What Do You Think?” (S#6). Relate the structures and their work to the worms’ survival and the possible effect of environmental agents on earthworm locomotion and survival.

2. Review the “Comparing Earthworm and Human Nervous Systems: Background Information” (C#4) that will introduce the concepts for the 3 experiments that follow (Days 7-10).

Segue
Discuss observations to date on how earthworms sense and respond to stimuli in their environment.

Tasks To Be Completed During the Class Period

- Introduce the concept of a model and remind students that the earthworm is being used as a model.
- Begin with “Comparing Earthworm and Human Nervous Systems” (C#4). (Discuss the concepts in this section that are most appropriate for the level and type of science class you are teaching.)
- Discuss how earthworms sense their environment and the function of the sensorimotor pathway.
- Discuss the lowest observed effect concentration (LOEC) of earthworm sensory neurons.
- Discuss how chemicals in the soil can stimulate a behavioral response by an earthworm.
- If time permits, segue to possible effects of environmental chemicals on burrowing behavior.
C#4 – COMPARING EARTHWORM AND HUMAN NERVOUS SYSTEMS

Introduction: Background Information

What is a model?

The characteristics of model organisms that have made them useful in research laboratories also make them well-suited to classrooms. Compared to general living organisms, model organisms are well-established experimental systems and are often ready to be used in classrooms. The available resources for these model organisms make the transition of their use to curricula relatively simple.

So, what exactly is a “model organism” and why are they so useful in the biology classroom? Model organisms are species that have basic biological processes common to many organisms, especially human. Due to their simplicity of structure and features, they are more amenable to asking certain questions. A model system is a simpler, idealized system that is generally accessible and easily manipulated. Model organisms: 1) have embryos or fetuses that develop rapidly and have short life cycles to quickly generate offspring, 2) have small adult sizes for ease of housing in a laboratory, 3) are readily available to allow for easy experimentation, and 4) are easily tractable, i.e., they have simpler systems so they are easier to experimentally manipulate.

There are four advantages to using model organisms in teaching biology. First, the model organism's immediate response to the change of environment will enhance students' learning and serve to hold their attention and interest. Students not only understand what they see, but relate more strongly to visual experiences. Second, the use of model organisms involves hands-on activities that provide a unique experience that could not be obtained with other teaching methods. Third, model organisms allow students to explore scientific methods and concepts themselves. Through this process, they will learn the investigative nature of science, including how to ask a question, collect data and draw conclusions. Fourth, students gain an appreciation that the biological insights gained from using model organisms have helped to improve our understanding of life in general and treat human diseases in particular.

Worms as a model for humans

Because the worm nervous system is much simpler than the human system, it is easier to study and, important for this module, manipulate to produce repeatable, stereotypic behavioral responses. The basic processes by which nerves work and stimulate muscles to contract are fundamentally the same as what occurs in humans (see Figure A). Therefore, observing neurobehavioral responses (movement caused by nerve-muscle interactions) provide insights into human behavior.
C#4 – COMPARING EARTHWORM AND HUMAN NERVOUS SYSTEMS

Figure A: Comparison of Earthworm and Human Nervous Systems

Above: Annelid Nervous System

Left: Human Nervous System
Source: http://www.newworldencyclopedia.org/entry/Nervous_system

Student Activity: Identify the relative location of each of the following in the worm and human nervous system: brain (central nervous system), source of nerves that connect to muscles, nerves that connect to the muscles (peripheral nervous system), and location of important sensory neurons.

How are the nervous systems in worms and humans similar? How are they different?

➢ Useful references on model organisms:

C#4 – COMPARING EARTHWORM AND HUMAN NERVOUS SYSTEMS

A good model organism should also be easy to use. Healthy worms are inexpensive and easy to obtain at any fish bait store. They are also easy to maintain in a classroom environment. In fact, vermiculture (the science of raising worms) is not only a good lesson in ecological relationships but it is big business. Here are some useful links:

How to raise worms:

List of companies in US that do vermiculture:
http://www.bae.ncsu.edu/topic/vermicomposting/vermiculture/directory-by-state.html

How do worms sense their environment?

Although we and all other animals really can sense many more stimuli (e.g., earth’s magnetic poles, polarized light, electrical signals), we typically think of there being 5 senses—sight, smell, sound, taste, and touch. Worms, like all animals, need to sense their environment to react appropriately to any changes that occur in their environment. Environmental stimuli are detected by sensory cells, sometimes called receptor cells, which detect dramatic and sometimes not so dramatic changes in their environment (in other words, receptor cells are really, really sensitive to very small changes in stimulus intensity). These cells may be scattered over the worm’s body or concentrated into a sensory organ. Sensory receptor cells are nerve cells specialized for transmitting information by transforming the energy of the external stimuli (e.g., touch) into an electrical impulse that passes down the neuron to some target tissue or organ.

What is the sensorimotor pathway?

Worms, like all animals, are able to navigate diverse and complex environments by transforming sensory cues into patterns of locomotion, e.g., alternating sequences of forward movements, reversals and turns. The navigation capabilities of human-made robots pale in comparison to the complexities of sensorimotor integration of even simple organisms. The earthworm finds its way in the mechanical, chemical and thermal conditions of the soil in which it finds itself using the properties of its neural circuitry. This circuit, called the sensorimotor pathway, consists of the sensory neuron, and the interneurons (nerve cells found within the central nervous system that link sensory and motor neurons) that connect, or synapse, onto motoneurons (sometimes called motor neurons, these nerves convey impulses toward or to muscles to make them contract and cause the animal to move). Frequencies and intensities of each of these behavioral components allow the worm to move forward or backward, and to change its direction of movement in response to specific environmental conditions, e.g., light intensity or presence of specific chemicals. Worms have different patterns of crawling in the presence or absence of bacterial food (worms don’t “eat dirt,” they ingest soil to digest the bacteria on the soil particles). If food is taken away, worms begin investigating the local vicinity with bursts of forward movement in various directions, frequent long reversals and turns, and suppressed short reversals. This is called area-restricted search behavior. But if food is still not found after a few minutes, worms switch to long-range roaming by suppressing all types of reorientation and crawling long distances with persistent forward movements. These exploratory movement patterns are distinct from the directed movement towards specific chemical or thermal cues (cues presumably connected with the presence of food or safety).
STUDENT ACTIVITY (OPTIONAL): Compare the movements of worms on paper towel, on sand and on potting soil. How do the movements compare, i.e., describe the area-restricted search behavior in each environment? Based on those movements and the description above, which do you think has more worm food? Why?

How do worms know where to burrow?

Using their sensory neurons for touch, vibrations, chemicals and light, worms test their environment to identify a good soil type into which to begin burrowing. Aspects of their environment which are important to them are: moisture level, density of soil (i.e., how easy is it to burrow into the soil), food availability (they eat bacteria so they are looking for items upon which bacteria will grow (e.g., food scraps), or presence of potential predators. Can you think of other potential environmental cues that might be important for the worm?

Simple diagram of sensorimotor pathway

Below are two illustrations of the worm nervous system. Note how the prostomium (the front-most lobe which is not a true segment) has a rich bed of sensory neurons (Figure B). Why would so many sensory neurons be located in the prostomium? In the micrograph (Figure C), notice how the sensory neurons connect to the segmental nerves, which then connect to the muscles. Why wouldn’t the sensory neurons connect directly to the muscles?

Figure B: Organization of the Sensory System of an Earthworm

The anterior part of the central nervous system of *Lumbricus* (side-view)

Source: http://cronodon.com/BioTech/Earthworm_NS.html

Note: Segmental nerves above are shown as green lines above but as red lines II and III in Figure C below.
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

C#4 – COMPARING EARTHWORM AND HUMAN NERVOUS SYSTEMS

Figure C: A 3-D Confocal Micrograph of Earthworm Sensory Elements

A 3D confocal micrograph of sensory elements at the border of two neighboring segments in an earthworm. In this view, you are looking at the dorsal surface of the worm, i.e., you are looking down on the worm with the left being toward the worm’s head (anterior) and the right toward its tail (posterior). The anatomical organization of the stained primary sensory cells (small white arrows at left of photograph) and their processes near the epithelial surface II., III., segmental nerves; A-P (upper left corner) = anterior–posterior direction.


Worm sensorimotor function

Photoreception

Earthworms have no eyes (although many other worms do, eyes are not so useful when you spend most of your life buried in soil or crawling about at night). However, worms do have light sensors in the form of specialized nerve endings. These sensors, or photoreceptor cells (see Figure D), occur in most parts of the worm's skin (epidermis) below the cuticle. They are more concentrated on the back and sides of the worm, although a few do occur on the ventral surface at the front end (1st segment) of the worm. They are also much more concentrated toward the front end of the worm, especially in the prostomium. There are also quite a few on the rear-most segment of the worm, including its ventral surface. Why there should be photoreceptors in the tail region?

Figure D: Earthworm Photoreceptors

About as simple as a photoreceptor cell can be, it contains a pigment that is sensitive to light which reversibly changes the shape of the pigment molecules. When the pigment molecules change shape, electrical changes in the receptor cell membrane occur that in turn lead to the propagation of a nerve signal.

The prostomium (Figure E) is innervated by a pair of branching nerves, of which some end in photoreceptors. Many of the larger branches have bulbous clusters of photoreceptors part-way along their length (Figure F). Remember that the prostomium is not very large so some light shines all the way through it in bright sunlight. It is believed that by comparing the signals coming from the subepidermal photoreceptors with those coming from the more shaded photoreceptors deeper within the prostomium (Figure G) the worm can gauge the light intensity.

**Figure E: Longitudinal Section of the Earthworm Prostomium**

Source: http://cronodon.com/BioTech/Earthworm_NS.html
A longitudinal section through the prostomium, showing the prostomial photoreceptors (nearer to the surface) and the deeper clusters of photoreceptors and the sub-epidermal nerve plexus.

A cross-section (transverse section) through the prostomium showing the photoreceptors, nerves and muscle cells. Notice how the epidermal sensor units are strategically placed along the sides of the prostomium.
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

C#4 – COMPARING EARTHWORM AND HUMAN NERVOUS SYSTEMS

• Color sensitivity: what and why

Earthworms will avoid light and strong sources of heat, including sunlight because their normal environment (soil and underground) is devoid of light and tends to be cooler. In your experiment, you will examine the effect of different colors of light on earthworm movement. **Why would earthworms respond differently to each color of visible light? Devise a testable hypothesis as to how they could detect these colors?** Furthermore, earthworms also react to strong, toxic chemicals and vibrations.

➢ Chemoreception

Chemoreception is the identification of chemical substances and their concentrations. This sensory mechanism is found in even the most primitive life forms. The mechanism is not completely understood, but it is known that receptor sites on cell membranes recognize specific molecules. Whether this occurs by chemical reaction, molecular shape, or some combination of the two is not known.

Figure H: Chemoreceptor Cells in an Earthworm

![Chemoreceptor Cells in an Earthworm](http://www.transtutors.com/biology-homework-help/earthworm/earthworm/behavior/)

Figure I: Relative Location of Chemoreceptor Cells in an Earthworm

![Relative Location of Chemoreceptor Cells in an Earthworm](http://cronodon.com/BioTech/Earthworm_NS.html)

Sensors in the epidermis consist of clusters of epidermal cells bearing cilia. These sensors are found scattered around the earthworm and may be touch sensors or chemoreceptors.


SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

C#4 — COMPARING EARTHWORM AND HUMAN NERVOUS SYSTEMS

The nervous system allows the animal to quickly detect, communicate and coordinate information about its external and internal environment so it can make efficient and appropriate responses for survival and/or reproduction. Sensory neurons carry electrical signals (impulses) from receptors or sense organs to the central nervous system (CNS). Sensory neurons are also called afferent neurons. The cell body of sensory neurons is outside the CNS in ganglia.

Earthworms, soil type and sensitivity to chemicals

➢ Thermoreception

Thermoreception is the detection of temperature changes and is present in most animals, including worms. Thermoreceptors in worms may also be sensitive to touch.

Figure J: Thermoreceptor Cells in an Earthworm

In the winter the earthworm travels deep in the soil to avoid the frost. In the hot summer the worm also travels deep into the soil in order to avoid dehydration. During the night an earthworm surfaces to let off their castings, an organic form of fertilizer produced from earthworms that is essentially earthworm waste. So, if it is dark and the worm is deep in the soil, how does it know it is nighttime?

Some earthworm species cannot tolerate low temperature. As a result, their “native” habitat is your compost bin. However, other earthworm species can tolerate low temperatures and are sometimes used for cold-weather composting. The risk is that, if they escape into the wild, they can populate areas in colder climates that previously were worm-free, areas where they have no natural competitors or predators.

➢ Connecting sensory neurons to muscle

Once a sensory neuron has detected a stimulus, it is important that this information gets to the muscles so the worm can move appropriately. Receptors detect the specific stimulus to which those neural endings are sensitive (Figure K, top). The message is sent down the axon to the synaptic endings that connect to the next neuron (blue arrow). In the case of the worms, this will
be the segmental nerves. These nerves connect to the muscle and send a message to contract (Figure K, bottom). The strength of the original stimulus determines how many motor neurons will be called into action, i.e., how strong the response to the stimulus will be.

**Figure K: A Simple Sensorimotor Nerve Circuit**

**Sensory Neuron or Afferent Neuron:** Moving away from a central organ or sensory neuron (red arrow). Relays messages from a stimulus (e.g., sun) from the receptors to the brain or spinal cord.

**Motor Neuron or Efferent Neuron:** Moving toward a central organ or point (green arrow). Relays messages from the brain or spinal cord to the muscles or other organs.

❖ What is LOEC?

The *lowest observed effect concentration (LOEC)* is the lowest concentration of a specific substance found by experiment or observation that causes an adverse change in morphology, function, growth, development, or lifespan of the exposed *population of organisms* (Why NOT of an individual?) under defined conditions of exposure. Federal agencies use LOEC to set chemical safety standards. The LOEC level does NOT mean that there is no affect below this concentration. It only means that for the given variable being examined under the specific
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

C#4 – COMPARING EARTHWORM AND HUMAN NERVOUS SYSTEMS

conditions to which the animal is being exposed, no effect could be detected with the instruments being used. Other variables could show an effect at lower levels or the same variable under different experimental conditions could result in a different (higher or lower) LOEC value. In this module, we are examining the LOEC of sensory neurons. Here are some links to research papers using LOEC to evaluate the effects of specific contaminants on earthworms.

Copper, lead, zinc: http://www.ncbi.nlm.nih.gov/pubmed/15091707

Soil type and ion reactivity—the concept of cation exchange capacity

Worms love soil. So to understand worm behavior and their reactions to chemicals in the soil, we need to first understand a little about how soil interacts with those chemical contaminants. For inorganic soils, the size of the soil particle is very important. Mineral soils are formed by the breakdown of rocks by chemical erosion, by weathering from heating and cooling, freezing and thawing, wind and water erosion, or biological activity into finer and finer particles. Eventually the particles get so small that they become colloidal clay, made up mostly of silica and alumina. A colloid is a particle that is so small that it remains suspended in water without becoming dissolved. The smaller a soil particle, the larger its surface area becomes relative to its size. Therefore, because colloids are so small, they have a very large surface area per unit volume or by weight. To visualize this, do the following exercise.

**Student Activity:** For each cube calculate the: 1) volume and 2) total surface area. Now calculate the ratio between surface area and volume.

**Figure L: Calculating the Surface Area/Volume Ratio of a Cube**

This difference in surface-to-volume ratio results in a greater concentration of negative charges on the surface of small particles vs. larger ones (Figures L[above] and M [below]). And, of
course, negative charges (anions) attract positive charges (cations). So, clay and humus act like magnets pulling cations from the water that exists between the soil particles. These minerals can then be replaced or exchanged by other cations (i.e., cation exchange). Some of these cations may bind tightly to the soil and are not easily leached when it rains. This ability of soil particles to hold positively charged ions is called cation exchange capacity (CEC).

Figure M: A Comparison between Two Types of Mineral Soil Particles
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

C#4 – COMPARING EARTHWORM AND HUMAN NERVOUS SYSTEMS

The size of the particles is also important in soils with organic matter mixed into it. Organic matter, as it breaks down, also forms smaller and smaller particles, until it breaks down as far as it can go and still be organic matter. At that stage it is called humus, and humus is also a colloid. (See Figure L above to learn why the ratio between surface area of a particle and its total volume are so important.) Some clays, such as vermiculite (the shiny particles often found in potting soil to help retain soil moisture), have a surface area as high as 800 square meters per gram, over 200,000 square feet (almost five acres) per ounce! The surface area of fully developed humus is about the same or even higher. So, the amount of humus, and the amount and type of clay, determine how much cation exchange capacity a given soil has (Table 1).

Table 1: Examples of CEC values for different soil textures

<table>
<thead>
<tr>
<th>Soil texture</th>
<th>CEC (meq/100g soil)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sands (light-colored)</td>
<td>3-5</td>
</tr>
<tr>
<td>Sands (dark-colored)</td>
<td>10-20</td>
</tr>
<tr>
<td>Loams</td>
<td>10-15</td>
</tr>
<tr>
<td>Silt loams</td>
<td>15-25</td>
</tr>
<tr>
<td>Clay and clay loams</td>
<td>20-50</td>
</tr>
<tr>
<td>Organic soils</td>
<td>50-100</td>
</tr>
</tbody>
</table>

CLASS DISCUSSION: Consider how all this can affect the potency of chemicals in the soil to stimulate a behavioral response by the earthworm.

❖ Metals as micronutrients and as toxic chemicals

Micronutrients are components of the diet that are required in relatively small quantities. In humans, deficiencies of these elements cause specific diseases, such as anemia (low Fe), decreased immune function (low Zn), and osteoporosis (low Ca). We know that 29 of the 90 naturally occurring elements are essential for life. Living matter consists of six basic structural elements (C, H, N, O, P and S), five macroelements (a chemical element required in relatively large quantities for the normal physiologic processes of the body, such as Ca, Mg, Na, K and Cl), and fifteen trace elements (such as Fe, Mn, Cu, Zn, Se, I, Co, Cr, Mo, F). However, if the micronutrients are present in the body at concentrations above what is needed, they can be toxic. An excellent discussion of this can be found at:

http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2246629/

In this module, we will be testing sensitivity to very high levels of some of these micronutrients, concentrations much higher than required for survival, concentrations that may even be toxic to earthworms.
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

C#4 – COMPARING EARTHWORM AND HUMAN NERVOUS SYSTEMS

How can soil become acidic?

Acid Rain

Acid rain is the wet and/or dry deposition (see explanation of these terms below) from the atmosphere containing higher than normal amounts of nitric and sulfuric acids. Acid rain can form from both natural sources, such as volcanoes and decaying vegetation, and man-made sources, primarily emissions of sulfur dioxide \((SO_2)\) and nitrogen oxides \((NO_x)\) resulting from fossil fuel combustion. In the United States, roughly \(2/3\) of all \(SO_2\) and \(1/4\) of all \(NO_x\) come from electric power generation that relies on burning fossil fuels, like coal. When these gases react in the atmosphere with water, oxygen, and other chemicals, they form a mild solution of sulfuric acid and nitric acid. The \(pH\) (a measure of hydrogen ion concentration in which the more hydrogen ions present in a solution, the more acid and less alkaline that solution is) of acid rain is often as low as 4. For that reason, this module will be examining the effect on worm behavior at \(pH\) 4. When sulfur dioxide and nitrogen oxides are released from power plants and other sources, prevailing winds blow these compounds across state and national borders, sometimes over hundreds of miles.

- **WET DEPOSITION**

Wet deposition refers to acidic precipitation in the form of rain, fog, or snow. As this acidic water flows over and through the ground, it affects a variety of plants and animals. The strength of the effects depends on several factors, including how acidic the water is; the chemistry and buffering capacity of the soils involved; and the types of fish, trees, and other living things that rely on the water. Soils with high amounts of limestone can buffer acid rain better than soils that are, for example, largely derived from granite or quartz.

- **DRY DEPOSITION**

Acid chemicals also may become adsorbed (or stuck to) onto dust or smoke and fall to the ground or on buildings, homes, cars, and trees through dry deposition. Dry deposited gases and particles can be washed from these surfaces by rainstorms, leading to increased acidic runoff (Figure N). About half of the acidity in the atmosphere falls back to earth through dry deposition.

Figure N: Sources of Acid Rain

SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

C#4 – COMPARING EARTHWORM AND HUMAN NERVOUS SYSTEMS

An excellent, short You Tube presentation on coal combustion and acid rain can be found at: http://www.britannica.com/eb/article-9003549/acid-rain?source=youtube_18494.

A good summary of the effects of acid rain can be found at: http://www.adirondackcouncil.org/acrapub.pdf.

B. Acid Mine Drainage

In many states, such as Wisconsin, mining activity has been an important part of the local economy. Metal ores are often associated with sulfide anions. As the ore is mined, these sulfides react with water to produce sulfuric acid (Figure O). This strong acid leaches into the streams and lakes of the watershed, acidifies those waterways, and releases potentially toxic metals into the water. This process is called acid mine drainage. Note that in Figure O, sulfuric acid, H2SO4, is written as SO4^2- + H^+ because in water (H2O), the acid breaks up into its reactive ionic parts.

Figure O: Chemical Reaction of Metal Sulfide Ores with Water to Form Sulfuric Acid

![Chemical Reaction of Metal Sulfide Ores with Water to Form Sulfuric Acid](source: Professor Mark Williams at the University of Colorado Boulder)

\[
4FeS_2 + 14 H_2 O + 15 O_2 \rightarrow 4Fe(OH)_3 + 8 SO_4^{2-} + 16 H^+
\]

Most organisms live within a well-defined range of pH values. If the pH falls below this tolerance range, death will occur due to respiratory or osmoregulatory (osmoregulation is the regulation of osmotic pressure especially in the body of a living organism) failure by disturbing the balance of sodium and chloride ions in the blood. At low pH, hydrogen ions may be taken into cells and sodium ions, necessary for proper cell function, are expelled.

Acid waters typically have fewer species and a lower abundance and biomass of macroinvertebrates than near-neutral pH waters. Low pH tends to eliminate species that feed on algae (scrapers or grazers such as snails). Low pH may inhibit growth of bacteria that help break down leaves into more easily digestible food for other aquatic animals.
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

C#4 – COMPARING EARTHWORM AND HUMAN NERVOUS SYSTEMS

Fish species are severely impacted at pH 4.5 to 5.5 due to loss of sodium ions from the blood and loss of oxygen in the tissues. Acid water also increases the permeability of fish gills to water, adversely affecting gill function. Ionic imbalance in fish may begin at a pH of 5.5 or higher, depending on the tolerance of the species; severe anoxia (low dissolved oxygen in the water) will occur below pH 4. Low pH that is not directly lethal may adversely affect fish growth rates and reproduction.

Heavy metals can increase the toxicity of mine drainage. Iron, aluminum, and manganese are the most common heavy metals which can compound the adverse effects of mine drainage. Heavy metals are generally less toxic at near-neutral pH. Trace metals such as zinc, cadmium, and copper (two of which are being tested in this module), which may also be present in mine drainage, are toxic at extremely low concentrations and may act synergistically to suppress algal growth and affect fish and benthic organisms. Some fish, such as brook trout, are tolerant of low pH, but the addition of metals decreases that tolerance.

Photo montage of streams affected by acid mine drainage. The orange color is due to iron hydroxide precipitates in the water (see chemical equation above).

- Relationship to cation exchange capacity (CEC)

In general, the CEC of most soils decreases with decreasing pH, i.e., highly acidic soils have less ability to bind cations than alkaline soils. This means that acidifying the soil makes metals, including toxic metals such as lead, cadmium, and aluminum, more available to plants and animals. Just how available is what this module will be examining.
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

CLASS DAY 7 – EXPERIMENT 1 – SIMPLE SENSORIMOTOR REACTIONS

Teacher Preparation for Class
1. Review “Experiment 1 – Simple Sensorimotor Reactions” (S#7)
3. Review “Ant Farm Set-up” video supplement and guide.
4. Review “Worms: Experiment #1” video supplement and guide.
5. Set out materials for each group.

Tasks To Be Completed During the Class Period
- Briefly review what students have learned about earthworm movement and the sensorimotor pathway.
- Demonstrate to students how to make the test chamber.
- Introduce Experiment 1 – Simple Sensorimotor Reactions

Materials

WORKSHEET:
“Experiment 1 - Simple Sensorimotor Reactions Data Sheet” (S#7)

FOR EACH GROUP:
- 1 small ant farm
- 1 container red worms (approx. 36 worms/container)
- Test tube rack
- 6 test tubes (10 mL) per solution tested to create serial dilutions of 1000, 100, 10, 1, 0.1, 0.01 ppm + a separate tube with dechlorinated water (0 ppm)
- Transfer pipets
- Forceps
- 1 squirt bottle and 1 liter beaker of dechlorinated water for rinsing worms between trials

Methods
Each of the 3 experiments are conducted on separate days except with Experiment 3 requiring two days unless the class is divided up so that some groups use sand and others use potting soil.
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

Solutions (2 L/class):

- 2-liter clean soda bottles
- pH 4, 7 and 9 (use the strong acids H$_2$SO$_4$ or HNO$_3$ as models of acid rain—HCl works, too)
- Optional: Vinegar (a weak acid: use full strength and a 10% and 1% solution)
- 1000 parts per million (ppm) of: Copper, Manganese, Nickel, and/or Zinc

Students are free to identify other chemicals of interest for these experiments.

Making Stock Solution and Serial Dilutions

The first step in making a serial dilution is to create a 1 liter stock solution of 1000 parts per million (ppm) metal salt as shown in Figure P using copper as an example. Take 1 mL of the stock solution and place it into 9 mL of distilled water (Why distilled water and not tap water?). This produces 10 mL of the dilute solution with 1 mL of extract /10 mL, producing a 10-fold dilution.

To make additional dilutions, the process is repeated sequentially (Figure P) using more and more dilute solutions as the "stock" solution. At each step, 1 mL of the previous dilution is added to 9 mL of distilled water. Each step results in a further 10-fold decrease in the concentration from the previous concentration. Review “Serial Dilutions” video supplement and guide.

Figure P: Method for Preparing a Serial Dilution from a Stock Solution

2.09 g CuCl$_2$

MW Cu$^{2+}$ = 64; MW Cl$^-$ = 35; MW CuCl$_2$ = 134

% Cu$^{2+}$ in CuCl$_2$ = 64/134 x 100% = 48%

1 ppm = 1 mg/L; 1000 ppm = 1000 mg/L = 1 g/L

Amount CuCl$_2$ to give 1000 mg Cu$^{2+}$ = 1000 x (134/64) = 2090 mg

2090 mg CuCl$_2$/L H$_2$O = 2.09 g CuCl$_2$/L H$_2$O = 1000 ppm Cu$^{2+}$
Making Test Chambers

Remove green sides of ant farm and separate clear plastic panels. Remove the plastic farm model. Place the panels on the table so that the insides are facing up. You now have 4 “raceways” for the worms. Review “Ant Farm Set-up” video supplement and guide.
S#7 – EXPERIMENT 1 – SIMPLE SENSORIMOTOR REACTIONS

PROCEDURE:

Note: After each worm is used for a trial, rinse it in clean, dechlorinated water in a squirt bottle and place it in a beaker of moist soil. By rinsing the worm’s surface of acid or metal solution, the worms can be reused for the next set of experiments, as well as for the next class.

A. Effect of pH: With a transfer pipet, create a circle of drops of pH 7 (dechlorinated water or distilled water) on the plastic raceway that is large enough to encircle your worm.

Why should you use dechlorinated water rather than water directly from your tap? Place worm on the raceway within the circle of drops. On your data sheet record the worm’s reaction (Strong, Mild, or No Reaction) when it comes into contact with the solution. Repeat for a total of 4 worms. When finished, wash the plastic raceway and dry with a paper towel. Repeat this procedure for pH 9 and 4.

OPTIONAL EXPERIMENTS:

1. Use additional solutions of pH 5, 6, 8 and 10 to create a more complete picture of the worms’ sensitivity to acids and bases.
2. Compare the reactions between a strong acid (HCl or H₂SO₄) vs. a weak acid (acetic acid/vinegar) at pH 4, 5 and 6.

B. Effect of Metals: Make all your serial dilutions of each of the 4 metal solutions as described above. Label each test tube with the metal concentration, e.g., 1000 ppm Cu, 100 ppm Cu, 10 ppm Cu, etc. Starting with the HIGHEST concentration first, repeat methods outlined in part A above for a total of 4 different worms/concentration. Rinse the raceway between each change in concentration. Record your observations on your data sheet. Once the worm has stopped reacting to the metal solution, you do not need to test lower concentrations. Metals should be chosen such that all four metals are tested by the class but any one group does 1-2 metals.
S#7 – EXPERIMENT 1 – SIMPLE SENSORIMOTOR REACTIONS DATA SHEET

Date:_______________  Hour:____________________   Acid: _______________________
Observers: ____________________________________________________________________

A. Effect of pH

<table>
<thead>
<tr>
<th>pH</th>
<th>Worm #</th>
<th>Strong Reaction</th>
<th>Mild Reaction</th>
<th>No Reaction</th>
</tr>
</thead>
<tbody>
<tr>
<td>4</td>
<td>1</td>
<td></td>
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<td>2</td>
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<td>4</td>
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<td>7</td>
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<td></td>
<td>4</td>
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<td>9</td>
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<td></td>
<td>4</td>
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</tr>
</tbody>
</table>

LOEC = Lowest Observed Effect Concentration

LOEC value for __________ acid is between pH __________ and __________.

OPTIONAL: Be an Environmental Toxicologist!
- Develop and conduct an experiment to find a more exact LOEC value!
- Compare the effects due to a strong acid vs. weak acid.
**S#7 – EXPERIMENT 1 – SIMPLE SENSORIMOTOR REACTIONS DATA SHEET**

Date: _______________       Hour: _______________

Observers: ____________________________________________________________________

**B. Effect of (Name of First Metal Tested):________________________________________**

<table>
<thead>
<tr>
<th>Concentration (ppm)</th>
<th>Worm #</th>
<th>Strong Reaction</th>
<th>Mild Reaction</th>
<th>No Reaction</th>
</tr>
</thead>
<tbody>
<tr>
<td>1000</td>
<td>1</td>
<td></td>
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<td>4</td>
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<td>100</td>
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<td>0.01</td>
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<tr>
<td>0</td>
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<td>4</td>
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</tbody>
</table>

**LOEC = Lowest Observed Effect Concentration**

LOEC value for ____________ is between __________ and __________ parts per million (ppm)
A. Effect of (Name of Second Metal Tested):______________________________

<table>
<thead>
<tr>
<th>Concentration (ppm)</th>
<th>Worm #</th>
<th>Strong Reaction</th>
<th>Mild Reaction</th>
<th>No Reaction</th>
</tr>
</thead>
<tbody>
<tr>
<td>1000</td>
<td></td>
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<td></td>
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<tr>
<td>100</td>
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<td>10</td>
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<tr>
<td>1.0</td>
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<tr>
<td>0.1</td>
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<tr>
<td>0.01</td>
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<tr>
<td>0</td>
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</tbody>
</table>

LOEC = **Lowest Observed Effect Concentration**

LOEC value for __________ is between __________ and __________ parts per million (ppm)
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

CLASS DAY 8 – EXPERIMENT 2 – PREFERENCE AVOIDANCE BEHAVIOR

Teacher Preparation for Class

1. Review “Experiment 2 – Preference Avoidance Behavior” (S#8)
2. Review “Worms: Experiment #2” video supplement and guide.
3. Set out materials for each group.

Tasks To Be Completed During the Class Period

- Briefly review what students have learned about simple sensorimotor reactions from Experiment 1.
- Introduce Experiment 2 – Preference Avoidance Behavior.

Materials

WORKSHEET:

“Experiment 2 – Preference Avoidance Behavior” (S#8)

FOR EACH GROUP:

1 small ant farm
1 container red worms (approx. 36 worms/container)
Test tube rack
6 test tubes (10 mL) per solution tested to create serial dilutions of 1000, 100, 10, 1, 0.1, 0.01 ppm + a separate tube with dechlorinated water (0 ppm)
Transfer pipets
Forceps
1 squirt bottle and 1 liter beaker of dechlorinated water for rinsing worms between trials
100 pre-cut paper towels (5 cm x 5 cm)

Solutions (2 L/class):

pH 4, 7 and 9 (use the strong acids H₂SO₄ or HNO₃ as models of acid rain—HCl works, too)
Optional: Vinegar (a weak acid: use full strength and a 10% and 1% solution)
1000 parts per million (ppm) of: Copper, Manganese, Nickel, and/or Zinc
S#8 – EXPERIMENT 2 – PREFERENCE AVOIDANCE BEHAVIOR

EXPERIMENT 2A: PROCEDURE

In this experiment, the worm can choose between different pH solutions, and then different concentrations of a metal solution to determine which side it will prefer.

Note: After each worm is used for a trial, rinse it in clean, dechlorinated water in a squirt bottle and place it in a beaker of moist soil. By rinsing the worm’s surface of acid or metal solution, the worms can be reused for the next set of experiments, as well as for the next class.

A. Effect of pH: With a transfer pipet, create 2 lines of drops as in the figure below, one of pH 7 (dechlorinated water or distilled water) AND one of pH 4 or 9 on the plastic raceway that is large enough to encircle your worm. Why should you use dechlorinated water rather than water directly from your tap?

Place worm on the raceway between the line of drops.

B. On your data sheet record the worm’s reaction (Strong, Mild, or No Reaction) when it comes into contact with the solution. Does it “prefer” one pH over the other? Repeat for a total of 4 worms. When finished, wash the plastic raceway and dry with a paper towel. Repeat this procedure for but use the other pH solution.

C. Effect of Metals: Make all your serial dilutions of the each of the 4 metal solutions as described above. Label each test tube with the metal concentration, e.g., 1000 ppm Cu, 100 ppm Cu, 10 ppm Cu, etc. Starting with the HIGHEST concentration first, repeat methods outlined in parts A and B above for a total of 4 worms/concentration. Once the worms seem to be choosing either side at random, i.e., they don’t seem to be avoiding or preferring one side over the other, you do not need to test lower concentrations. Rinse the raceway between each change in concentration. Record your
observations on your data sheet. Metals should be chosen such that all four metals are tested by the class but any one group does 1-2 metals.

EXPERIMENT 2B: PROCEDURE

In this experiment, the worm can choose between distilled water and a particular solution of a chemical. In other words, will the worm sense which side it will prefer.

PROCEDURE:

A. Place 1 pre-cut paper towel at each end of the raceway. With a transfer pipet, moisten (not soak) one towel with dechlorinated or distilled water (Figure Q). You can use the same worms from Experiment 1.

B. **Effect of pH:** With a transfer pipet, moisten (not soak) the paper towel on the other end of the raceway with pH 9. Place the worm on the bare plastic between the two towels. Record on your data sheet which side it chooses. Repeat for a total of 4 worms. Dispose of the towels into the trash, rinse and dry the raceway. Use same procedure for pH 4 and vinegar.

C. **Effect of Metals:** Make all your serial dilutions of the each of the 4 metal solutions as described above. Label each test tube with the metal concentration, e.g., 1000 ppm Cu, 100 ppm Cu, 10 ppm Cu, etc. Starting with the **HIGHEST** concentration first, repeat methods outlined in parts A and B above for a total of 4 worms/concentration. **Once the worm has stopped reacting to the metal solution, you do not need to test lower concentrations.** Rinse the raceway between each change in concentration. Record your observations on your data sheet. Metals should be chosen such that all four metals are tested by the class but any one group does 1-2 metals.
Figure Q: Diagrammatic View for Experiment 2B Methods
## EXPERIMENT 2A: PROCEDURE

### A. Effect of pH

For each row, place a check in the box labeled # Observations each time you observe a worm choosing a specific pH value among the choices given in that row. Enter the number of total check marks for that pH choice in the box labeled Total Observations. An example is provided in the row labeled Sample Recording of Data.

<table>
<thead>
<tr>
<th>pH Choice</th>
<th># Worms Found In:</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>pH 4</td>
<td>pH 7</td>
<td>pH 9</td>
<td></td>
</tr>
<tr>
<td></td>
<td># Observations</td>
<td>Total Observations</td>
<td># Observations</td>
<td>Total Observations</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
<td>/</td>
<td>1</td>
<td>///</td>
<td>3</td>
</tr>
<tr>
<td>pH 4 vs. pH 7</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>pH 9 vs. pH 7</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### EXPERIMENT 2A: PROCEDURE (continued)

#### B. Effect of Metal Concentration

Name of Metal Tested: ___________________________________________

<table>
<thead>
<tr>
<th>Concentration (parts per million)</th>
<th># Worms Found in:</th>
<th>Control Side (Water)</th>
<th>Treated Side</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td># Observations</td>
<td>Total</td>
<td># Observations</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
<td>/</td>
<td>1</td>
<td>///</td>
</tr>
<tr>
<td>1000</td>
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<td>0</td>
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</tr>
</tbody>
</table>
EXPERIMENT 2B: PROCEDURE

A. Effect of pH

Acid Used: ______________________________

For each row, place a check in the box labeled # Observations each time you observe a worm choosing a specific pH value among the choices given in that row. Enter the number of total check marks for that pH choice in the box labeled Total Observations. An example is provided in the row labeled Sample Recording of Data.

<table>
<thead>
<tr>
<th>pH Choice</th>
<th># Worms Found In:</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>pH 4</td>
<td>pH 7</td>
<td>pH 9</td>
<td></td>
</tr>
<tr>
<td></td>
<td># Observations</td>
<td>Total</td>
<td># Observations</td>
<td>Total</td>
</tr>
<tr>
<td>pH 4 vs. pH 7</td>
<td>/</td>
<td>1</td>
<td>///</td>
<td>3</td>
</tr>
<tr>
<td>pH 9 vs. pH 7</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
EXPERIMENT 2B: PROCEDURE (continued)

B. Effect of Metals

Name of First Metal Tested: __________________________________________

<table>
<thead>
<tr>
<th>Concentration of Treated Side (parts per million)</th>
<th># Worms Moving Toward:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control Side (Water)</td>
</tr>
<tr>
<td></td>
<td># Observations</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
<td>/</td>
</tr>
<tr>
<td>1000</td>
<td></td>
</tr>
<tr>
<td>100</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td></td>
</tr>
<tr>
<td>0.1</td>
<td></td>
</tr>
<tr>
<td>0.01</td>
<td></td>
</tr>
<tr>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

LOEC = Lowest Observed Effect Concentration

LOEC value for ___________ is between __________ and __________ parts per million (ppm)
Date: _______________       Hour: _______________

Observers: ________________________________________________________________

### B. Effect of Metals

Name of Second Metal Tested: ________________________________________________

<table>
<thead>
<tr>
<th>Concentration of Treated Side (parts per million)</th>
<th># Worms Moving Toward:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control Side (Water)</td>
</tr>
<tr>
<td></td>
<td># Observations</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
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</tr>
<tr>
<td>1000</td>
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<td>100</td>
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<tr>
<td>1</td>
<td></td>
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<tr>
<td>0.1</td>
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<tr>
<td>0.01</td>
<td></td>
</tr>
<tr>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

LOEC = **Lowest Observed Effect Concentration**

LOEC value for __________ is between ______ and _______ parts per million (ppm)
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

CLASS DAYS 9-10 – EXPERIMENT 3 – EFFECT OF SOIL TYPE ON BEHAVIORAL RESPONSES

Teacher Preparation for Class

1. Review “Experiment 3 – Effect of Soil Type on Behavioral Responses” (S#9)
2. Review “Worms: Experiment #3” video supplement and guide.
3. Set out materials for each group.
4. If you need to reduce the number of worms used for the entire class, divide the class into groups with each group doing a separate experiment. After all tests are completed, students should share their data to make a class graph.
5. Have the students collect 2 liter soda bottles. Rinse the bottles well and label each for the 3 pH values and for each of the metal concentrations. Since larger volumes of solution will be required to moisten the soil, these bottles will be used by all groups.

Tasks To Be Completed During the Class Period

- Briefly review what students have learned about preference avoidance behavior from Experiment 2.
- Conduct Experiment 3 – Effect of Soil Type on Behavioral Responses.

Materials

WORKSHEET:

“Experiment 3 – Effect of Soil Type on Behavioral Responses” (S#9)

FOR EACH GROUP:

1 small ant farm
1 container red worms (approx. 36 worms/container)
1 plastic divider to divide soil into two sections
Transfer pipets
Forceps
1 squirt bottle and 1 liter beaker of dechlorinated water for rinsing worms between trials
Test tube rack
6 test tubes (10 mL) per solution tested to create serial dilutions of 1000, 100, 10, 1, 0.1, 0.01 ppm + a separate tube with dechlorinated water (0 ppm)

Soil: Potting soil and sand

Treated soil container
### SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

**Solutions (2 L/class):**

<table>
<thead>
<tr>
<th>Item</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>31 2-liter clean soda bottles</td>
<td></td>
</tr>
<tr>
<td>pH 4, 7 and 9 (use the strong acids $H_2SO_4$ or $HNO_3$ as models of acid rain—HCl works, too)</td>
<td></td>
</tr>
<tr>
<td>Optional: Vinegar</td>
<td>(a weak acid: use full strength and a 10% and 1% solution)</td>
</tr>
<tr>
<td>1000 parts per million (ppm) of: Copper, Manganese, Nickel, and/or Zinc</td>
<td></td>
</tr>
</tbody>
</table>
S#9 – EXPERIMENT 3 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES

This experiment is similar to Experiment 2, except soil is used instead of paper towel.

PROCEDURE:

A. **Effect of Sand:** Into one raceway, add enough sand (sandbox sand, not the “sand” provided in the ant farm box) as shown in Figure R below to fully cover the plastic surface and to be at a depth equal to the thickness of the raceway. Place the plastic divider widthwise into the sand so that it divides the sand into two equal sections. Move the divider side to side to create a small gap (approx. 1-1.5 cm) between the two sand sections.

Figure R: Diagrammatic View for Experiment 3 Methods

Temporary barrier to: 1) prevent water and chemical solution from mixing, and 2) create space to place worm. Barrier removed prior to placing worm in raceway.
S9 – EXPERIMENT 3 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES

1. **Effect of pH:** Add \( \leq 30 \) mL of distilled water to the sand on one side of the divider. Sand should be moist to damp but not soaking. Add a similar amount of pH 9 to the sand on the other side of the divider. Place the worm in the gap and record which side it chooses. Repeat for 4 different worms. **Remove the pH 9 sand, dispose of it in a “Treated Soil” container, and wipe the section clean.** Add clean, dry sand and add a volume of pH 4 or vinegar (weak acid, i.e., is partially dissociated in an aqueous solution). Repeat procedure and record observations.

2. **Effect of Metals:** Remove all sand from previous experiment and replace with fresh, clean, dry sand after rinsing and drying the raceway. Make all your serial dilutions of each of the 4 metal solutions. **Label each test tube with the metal concentration, e.g., 1000 ppm Cu, 100 ppm Cu, 10 ppm Cu, etc. Starting with the HIGHEST concentration first, repeat methods outlined in part A.a above using clean, dry sand for each new concentration for a total of 4 different worms/concentration.** Once the worm has stopped reacting to the metal solution, you do not need to test lower concentrations. Metals should be chosen such that all four metals are tested by the class but any one group does 1-2 metals.

**B. Effect of Potting Soil:** Into one raceway, add enough potting soil as shown below to fully cover plastic surface and to be at a depth equal to the thickness of the raceway. Place the plastic divider widthwise into the potting soil so that it divides the potting soil into two equal sections. Repeat the methods above except you are now using potting soil instead of sand. You will likely need to use more water to make the potting soil moist-damp than you needed for sand.

![Potting soil](image)

**C. Disposal of Treated Soil:** Sand or potting soil that was treated with distilled water can be reused. Sand or potting soil that was treated with acid, base, or metal solutions cannot be reused. However, it can be placed in a compost pile (a good place to also place your worms after the experiments are completed!). When diluted with other soil in the bin, the metals provide good nutrients for future plants.
### A. Effect of pH

**Acid Used:**

For each row, place a check in the box labeled # Observations each time you observe a worm choosing a specific pH value among the choices given in that row. Enter the number of total check marks for that pH choice in the box labeled Total Observations. An example is provided in the row labeled Sample Recording of Data.

<table>
<thead>
<tr>
<th>pH Choice</th>
<th># Worms Found In:</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>pH 4</td>
<td>pH 7</td>
<td>pH 9</td>
<td></td>
</tr>
<tr>
<td></td>
<td># Observations</td>
<td>Total</td>
<td># Observations</td>
<td>Total</td>
</tr>
<tr>
<td>Sample</td>
<td></td>
<td>Observations</td>
<td></td>
<td>Observations</td>
</tr>
<tr>
<td>Recording</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>of Data</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>pH 4 vs. pH 7</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>pH 9 vs. pH 7</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
S#9 – EXPERIMENT 3 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES

Date: _______________       Hour: ____________________

Chemical: ________________________________   Soil Type: Sand

Observers: _______________________________________

<table>
<thead>
<tr>
<th>Concentration (parts per million)</th>
<th># Worms Found In:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control Side (Water)</td>
</tr>
<tr>
<td></td>
<td># Observations</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
<td>/</td>
</tr>
<tr>
<td>1000</td>
<td></td>
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<tr>
<td>100</td>
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<td>1</td>
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<tr>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

B. Effect of Metals

Name of First Metal Tested: _______________________________________

Chemical: ________________________________   Soil Type: Sand

<table>
<thead>
<tr>
<th>Concentration (parts per million)</th>
<th># Worms Found In:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control Side (Water)</td>
</tr>
<tr>
<td></td>
<td># Observations</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
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<td>1</td>
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<td>0</td>
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</tr>
</tbody>
</table>

Name of Second Metal Tested: _______________________________________

Chemical: ________________________________   Soil Type: Sand

<table>
<thead>
<tr>
<th>Concentration (parts per million)</th>
<th># Worms Found In:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control Side (Water)</td>
</tr>
<tr>
<td></td>
<td># Observations</td>
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<td>Sample Recording of Data</td>
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<td>1</td>
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<tr>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>
# Experiment 3 – Effect of Soil Type on Behavior Responses

Date: ___________________       Hour: ___________________

Chemical: ___________________________   Soil Type: **Potting Soil**

Observers: ____________________________________________________________________

## A. Effect of pH

**Acid Used:** __________________________

For each row, place a check in the box labeled **# Observations** each time you observe a worm choosing a specific pH value among the choices given in that row. Enter the number of total check marks for that pH choice in the box labeled **Total Observations**. An example is provided in the row labeled **Sample Recording of Data**.

<table>
<thead>
<tr>
<th>pH Choice</th>
<th>pH 4 Observations</th>
<th>Total Observations</th>
<th>pH 7 Observations</th>
<th>pH 9 Observations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample Recording of Data</td>
<td>/</td>
<td>1</td>
<td>///</td>
<td>3</td>
</tr>
<tr>
<td>pH 4 vs. pH 7</td>
<td></td>
<td></td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>pH 9 vs. pH 7</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### S#9 – EXPERIMENT 3 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES

Date: _______________       Hour: _________________

Chemical: ________________________________   Soil Type: **Potting Soil**

Observers:_____________________________________________________________________

### B. Effect of Metals

<table>
<thead>
<tr>
<th>Concentration (parts per million)</th>
<th># Worms Found In:</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control Side (Water)</td>
<td>Treated Side</td>
<td></td>
</tr>
<tr>
<td></td>
<td># Observations</td>
<td>Total</td>
<td># Observations</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
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<td>100</td>
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<td>10</td>
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<tr>
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<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Name of Second Metal Tested:____________________________________________

Chemical: ________________________________   Soil Type: **Potting Soil**

<table>
<thead>
<tr>
<th>Concentration (parts per million)</th>
<th># Worms Found In:</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control Side (Water)</td>
<td>Treated Side</td>
<td></td>
</tr>
<tr>
<td></td>
<td># Observations</td>
<td>Total</td>
<td># Observations</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
<td>/</td>
<td>1</td>
<td>///</td>
</tr>
<tr>
<td>1000</td>
<td></td>
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<td>100</td>
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<td>10</td>
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<tr>
<td>0</td>
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</tr>
</tbody>
</table>
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

CLASS DAYS 11-12 – EXPERIMENT 4 – EFFECT OF SOIL TYPE ON BEHAVIORAL RESPONSES

Teacher Preparation for Class

1. Review “Experiment 4 – Effect of Soil Type on Behavioral Responses” (S#10)
2. Set out materials for each group.
3. Prepare pH and metal solutions (preparation instructions below)
4. If you need to reduce the number of worms used for the entire class, divide the class into groups with each group doing a separate experiment. After all tests are completed, students should share their data to make a class graph.

Tasks To Be Completed During the Class Period

- Briefly review what students have learned about preference avoidance behavior from Experiment 3.
- Conduct Experiment 4 – Effect of Soil Type on Behavioral Responses.

Materials

WORKSHEET:

“Experiment 4 – Effect of Soil Type on Behavioral Responses” (S#10)

FOR EACH GROUP:

1 small ant farm
1 container red worms (approx. 36 worms/container)
1 plastic divider to divide soil into two sections
Transfer pipets
Forceps
1 squirt bottle and 1 liter beaker of dechlorinated water for rinsing worms between trials
Tape
pH paper
Ruler
Soil: Potting soil and sand
Treated soil container
Ring stand
Stop watch
pH 4 and 9 solutions (preparation directions below)
Instructions for making pH solutions

Making pH 4 from HCl

1. Dissolve 83.33ml of the concentrated HCL to water to make a 1 liter solution. This will produce pH = 0.0.

2. Using the stock solution from Step 1, you can now make a series of solutions with a scale of pH by successive 10 fold dilutions.
   • Dilute 100ml of the stock solution to 900 mL of water to make 1000ml of the 1M solution to get pH =1.0
   • Dilute 100ml of the pH=1.0 solution to 900 mL of water to make 1000ml solution of pH = 2.0
   • Dilute 100ml of the pH=2.0 solution to 900 mL of water to make 1000ml solution of pH = 3.0
   • And so on to get your desired pH 4, 5 and 6.

3. To get pH 4.0 directly, dilute 0.1 ml of the 1M solution and dilute to 1000ml to get pH=4.0

Making pH 9 from NaOH

1. Since pH 9 = pOH 5, OH\textsuperscript{-} concentration in pH 9 = 0.00001

2. Since concentration = grams/volume, grams = concentration x volume.

3. g OH\textsuperscript{-}needed = 0.00001 x 1000 mL solution = 0.01 g

4. Mass of NaOH needed = g OH\textsuperscript{-} needed x molecular mass of NaOH; 0.01 x 40 = 0.4 g NaOH needed in 1000 mL distilled water to make pH 9.
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

Measuring Soil pH
There are several ways that you can test the pH of your soil samples:

1. **Use the pH soil testing kit** provided or purchased from a home and garden center. Carefully follow all of the manufacturer’s directions. This is the most accurate test.

2. **Use pH Paper** When measuring pH with pH paper, dip the end of a strip of pH paper into each mixture you want to test. After about two seconds, remove the paper, and immediately compare the color at the wet end of the paper with the color chart provided with that pH indicator. Write down the pH value and color. Always use a clean, unused strip of pH paper for each mixture that you test.

*Remember the pH scale is based on a value of 7 representing neutral — that means neither acidic nor basic. Increasing values from 7.1 to 14 mean increasingly stronger bases. Decreasing values from 6.9 down to 0 mean increasingly stronger acids. Strong acids or bases are caustic, which means they will cause chemical burns! You are unlikely to find very strong acid or base values in soil — the plants would die.*

<table>
<thead>
<tr>
<th>Soil Sample</th>
<th>Soil pH or color of indicator</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td></td>
</tr>
</tbody>
</table>

**Changing the pH of Soil**
Two materials commonly used for lowering the soil pH are aluminum sulfate and sulfur. These can be found at a garden supply center. Aluminum sulfate will change the soil pH instantly because the aluminum produces the acidity as soon as it dissolves in the soil. Sulfur, however, requires some time for the conversion to sulfuric acid with the aid of soil bacteria and may be very slow (up to months). Both materials should be worked into the soil after application to be most effective.
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

<table>
<thead>
<tr>
<th>Present pH</th>
<th>Desired pH</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>6.5</td>
</tr>
<tr>
<td>8.0</td>
<td>1.8</td>
</tr>
<tr>
<td>7.5</td>
<td>1.2</td>
</tr>
<tr>
<td>7.0</td>
<td>0.6</td>
</tr>
<tr>
<td>6.5</td>
<td></td>
</tr>
<tr>
<td>6.0</td>
<td></td>
</tr>
</tbody>
</table>

*10 square feet is sufficient for ~36 ant farm set ups/pH choice. Use that as a guide for how much soil needs to be prepared in advance for each pH level to be tested and how much aluminum sulfate you will need.

Aluminum sulfate is toxic. Only the teacher should handle this chemical to prepare the soil pH. Students should wear gloves and, if available, lab coats while handling soil. Wash hands thoroughly after the experiment.

An alternative method that will involve the students is for the teacher to prepare a 1 L aluminum sulfate solution ahead of time (15 mL solid aluminum sulfate to 1 L distilled water will produce a pH 4 solution). Divide out 100 mL aliquots for the students. Students can then use those samples to pipet desired amount on the soil in their ant farm.

Aluminum sulfate is a chemical compound with the formula Al₂(SO₄)₃. It is soluble in water and is mainly used as a flocculating agent in the purification of drinking water and waste water treatment plants, and also in paper manufacturing. Landscapers will use it to reduce the pH (increase acidity) of the soil for specific acid-loving plants. Aluminum sulfate is sometimes referred to as a type of alum. The anhydrous form occurs naturally as a rare mineral found in volcanic environments and on burning coal-mining waste dumps. Aluminum sulfate, however, is usually found as a hydrated salt, e.g., hexadecahydrate Al₂(SO₄)₃•16H₂O and octadecahydrate Al₂(SO₄)₃•18H₂O.
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

Increasing the Soil pH
To increase soil pH, the common practice is to apply a material that contains some form of lime. Ground agricultural limestone is most often used. The finer the limestone particles, the more rapidly it becomes effective. Different soils will require a different amount of lime to adjust the soil pH value. The texture of the soil, organic matter content and the plants to be grown are all factors to consider in adjusting the pH value.

Selecting a Liming Material: You can choose from four types of ground limestone products: pulverized, granular, pelletized and hydrated. The finer the grind of the limestone the faster it will change the soil pH value.

Most liming materials are only slightly soluble in water, so incorporation in the soil is a must for lime reaction. Even when properly mixed with the soil, lime will have little effect on pH if the soil is dry. Moisture is essential for the lime-soil reaction to occur.

Wood Ashes: Wood ashes can be used to raise the soil pH. They contain fairly high amounts of potassium & calcium, and small amounts of phosphate, boron and other elements. They are not as effective as limestone but with repeated use, they can drastically raise the pH value of a soil, especially if the soil is sandy in texture. Coal ashes have no liming value and may actually lower pH. A good, easy source for wood ash is your fireplace residue.

Preparing the samples
Soil samples should be completely dry before testing. Open the bags and leave them in a sunny location until they are dry. Samples may also be dried in an oven. Spread a soil sample on a Teflon cookie sheet and place in a 350-degree oven for 15 minutes. Make sure the cookie sheet is cleaned and dried for each new sample.
S#10 – EXPERIMENT 4 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES

This experiment is similar to Experiment 3, except that the worm is burrowing first through a control soil before it encounters a treated soil. **BE SURE TO WEAR LAB GLOVES DURING THIS EXPERIMENT AS YOU WILL BE HANDLING CHEMICALS.**

**DISPOSAL:**
All used potting soil should be discarded into a specially designated bucket after all worms have been tested for each pH and metal concentration. Sand or potting soil that was treated with distilled water can be reused. Sand or potting soil that was treated with acid, base, or metal solutions cannot be reused. However, it can be placed in a compost pile (a good place to also place your worms after the experiments are completed!). When diluted with other soil in the bin, the metals provide good nutrients for future plants. Rinse and dry the ant farm between each experiment.

**PROCEDURE:** Refer to Figure S for a diagrammatic view for Experiment 4 methods.

**A. Effect of pH:**
1. Prepare a pH 4 solution using 15 mL of aluminum sulfate (Al₂(SO₄)₃) dissolved into 1 liter of distilled water. Use NaOH or KOH solution using distilled water to prepare pH 9. **Why not use dechlorinated water?**
2. Tape the divider that separates the two raceways so that the hole in the divider is blocked (Fig. S1). Tape the two pieces together so that the hole created when the two pieces are assembled is covered (Fig. S2a). Turn the assembly over and place a piece of masking tape 6 cm from the bottom (Fig. S2b). This will indicate to what level the bottom layer of will extend. Another piece of tape will be placed 10 cm above this mark and will indicate the volume for the top layer of soil. Mark the left side TREATED and the right side UNTREATED.
3. Into left raceway (TREATED), add approx. 15 ml of potting soil to the bottom half of the worm raceway (Fig. S3a). Keep it loosely packed so that it comes to a height of 6 cm from the bottom end of the raceway. If the soil is too tight, the worm will have difficulty burrowing and you will have difficulty seeing the worm in the raceway. Gently add 10 mL of either pH 4 or 9 to the soil. Be sure that it is evenly moist. Do the same procedure on the right half of the raceway (UNTREATED) except that you will add only distilled water. **WHY?**
4. Add 10 cm of potting soil to the top half of the raceway so that you now have 16 cm of soil (Fig. S3b). To this second layer of soil, add 5 mL of distilled water. Repeat on the other side of the worm raceway. **DO NOT LET THE TOP AND BOTTOM SOILS TOUCH EACH OTHER UNTIL BOTH HALVES HAVE BEEN TREATED WITH THEIR RESPECTIVE SOLUTIONS!! WHY?**
5. Using the pH soil test kit, measure the potting soil pH at 3 different locations on each side of the raceway before adding the worms and closing up the apparatus: bottom, top, and 1 cm above the location where the two soils meet. Record this information on Data Sheet A. **Why is this important?**
6. Place 2 worms onto the top layer of each raceway (2 on treated side and 2 on untreated side) and close up the ant farm. Clamp entire apparatus to a ring stand.
S#10 – EXPERIMENT 4 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES

7. Observe the worm movement over a 10 minute period. Record on Data Sheet B how far down (cm) the worm moved for each raceway.
8. Open the ant farm, remove the worms, and repeat procedure with 4 new worms. Continue until you have tested at least 6 worms for each pH.

B. Effect of Metals:
1. Procedure is the same as above except you will be using metal solutions.
2. Into left raceway (TREATED), add approx. 15 ml of potting soil to the bottom half of the worm raceway (Fig. S3a). Keep it loosely packed so that it comes to a height of 6 cm from the bottom end of the raceway. If the soil is too tight, the worm will have difficulty burrowing and you will have difficulty seeing the worm in the raceway. Gently add 5 mL of 1000 ppm metal solution to the soil. Be sure that it is evenly moist. Do the same procedure on the right half of the raceway (UNTREATED) except that you will add only distilled water. WHY?
3. Add 10 cm of potting soil to the top half of the raceway so that you now have 16 cm of potting soil. To this second layer of potting soil, add 5 mL of distilled water. Repeat on the other side of the worm raceway (Fig. S3b). DO NOT LET THE TOP AND BOTTOM SOILS TOUCH EACH OTHER UNTIL BOTH HALVES HAVE BEEN TREATED WITH THEIR RESPECTIVE SOLUTIONS!! WHY?
4. Place 2 worms onto the top layer of each raceway (2 on treated side and 2 on untreated side) and close up the ant farm. Clamp entire apparatus to a ring stand (Fig. S4).
5. Observe the worm movement over a 10 minute period. Record on Data Sheet B how far down the worm moved for each raceway.
6. Open the ant farm, remove the worms, and repeat procedure with 4 new worms. Continue until you have tested at least 6 worms for each treatment.
7. Repeat procedures (B2-B6) above except use a 100 ppm metal solution for the bottom layer on the treated side.
8. Again, repeat procedures (B2-B6) above except use 10 ppm and 1 ppm metal solutions.

C. Effect of soil type: Repeat entire protocol except use sand instead of potting soil.

NOTES:
1) To save class time, each pair of students can be assigned a specific comparison, e.g., 1000 and 100 ppm metal solution with potting soil. Have all students begin their experiment at the same time and then at the end of the 10-minute period, have students record their data on a common data form so all students can record the data from the entire class on their Data Sheet. In this way, over a 2-day period, the effect of pH, metals, and soil type can be observed and recorded by all the students.
2) If the worm has gone below 6 cm, it has entered the layer with the chemical contaminant. There may be some diffusion of the chemical into the top layer closest to the contaminated soil. If this happens, how would you determine this simply by observing the worm’s pattern of burrowing?
Optional Experiments: What other chemicals could contaminate your soil? How sensitive are worms to them?

Figure S:

1. Preparation of Worm Raceway

2. Marking Soil Levels on Worm Raceway
   a.  
   b. 

3. Adding Soil and Chemical Solutions
   a.  
   b. 

4. Assembled Apparatus

5. pH Soil Test Kit
<table>
<thead>
<tr>
<th>TREATED</th>
<th>UNTREATED</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH __________</td>
<td>pH __________</td>
</tr>
<tr>
<td>pH __________</td>
<td>pH __________</td>
</tr>
<tr>
<td>pH __________</td>
<td>pH __________</td>
</tr>
</tbody>
</table>

**S#10 – EXPERIMENT 4 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES-DATA SHEET A**

Date: ________________________   Hour: _________________________
pH: _________________________   Soil Type: ______________________
Observers: __________________________________________________________
**S#10 – EXPERIMENT 4 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES - DATA SHEET B**

Date: ________________________   Hour:  _________________________

Chemical: _________________________    Soil Type: ____________________________

Observers: ___________________________________________________________________

Name of metal tested: ___________________________________________________________________

<table>
<thead>
<tr>
<th>Worm #</th>
<th>Distance (cm) burrowed from top with bottom layer containing:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>pH 4</td>
</tr>
<tr>
<td>1</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td></td>
</tr>
<tr>
<td>Average</td>
<td></td>
</tr>
</tbody>
</table>

Observations:
Answer the following questions related to this experiment:

1. For each of the trials, why was it important to have one side of the raceway treated with only distilled water (labelled as “untreated”)?

___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
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___________________________________________________________________________

2. For the pH experiment, why was it important to measure the soil pH at 3 different locations on each side of the raceway before adding the worms and closing up the apparatus?

___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________

3. Was there a difference in burrowing depth for pH 4, pH 9, and the control side? If so, suggest a hypothesis for that difference.

___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________

4. Was there a difference in burrowing depth for 1000 ppm, 100 ppm, 10 ppm and 1 ppm of the metal solutions? If so, suggest a hypothesis for that difference.

___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________

5. Was there a difference in burrowing depth in sand vs. potting soil? If so, suggest a hypothesis for that difference.

___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

CLASS DAY 13 – OBSERVING CATION EXCHANGE CAPACITY OF SOILS

Teacher Preparation for Class
1. Review “Observing Cation Exchange Capacity of Soils” (S#11)
2. Set out materials for each group.

Tasks To Be Completed During the Class Period
- Briefly review what students have learned about the effect of soil types on behavior responses from Experiment 3.
- Introduce “Observing Cation Exchange Capacity of Soils” experiment.

Materials

WORKSHEET:
“Observing Cation Exchange Capacity of Soils” (S#11)
“Sensorimotor Reactions and Behavior Responses” – Assignment (S#12)

FOR EACH GROUP:
- Dissecting microscope
- 0.2% methylene blue solution
- Pipettes
- Potting soil
- Sand
- Balance (measures to 0.1 g)
- Weigh boats or paper
- Spatula
- Petri dishes
- Ruler (in centimeters)
- Distilled water
- 2 – 100 mL beakers (1 for potting soil, 1 for sand)
- Clear acetate transparency
- Fine tip marking pen
- 2 stop watches
- Concentrated metal solution (1000 parts per million) that has a strong color, e.g., nickel or copper
**S#11 – OBSERVING CATION EXCHANGE CAPACITY OF SOILS**

**PROCEDURE:**

A. By each balance, place 2 – 100 mL beakers, one of which will be filled 1/3 with potting soil and the other 1/3 with sand. Moisten the two soil types with distilled water but do not soak. Drain excess water.

B. On the acetate transparency, draw two 1 cm diameter circles with each circle 1 cm from the next circle. Place the petri dish over the two circles and place the petri dish and the transparency onto the dissecting scope stage. Adjust the focus so that you can see both circles simultaneously.

C. Weigh out 0.05g – 0.075 g of each soil type. Place each soil type in the Petri dish over one of the circles. Spread out so that the soil particles are as separated as possible but still within the 1 cm diameter circle. Repeat for the other soil type.

D. Add 1 drop of methylene blue to the potting soil and immediately start one of the stop watches. Then add 1 drop of methylene blue to the sand and immediately start the other stop watch. On the data sheet record the time (in seconds) for the blue color to visibly begin disappearing, and then the time at which all the blue color is gone for each soil type. Observe for a maximum of 5 minutes.

E. Repeat with any concentrated metal solution (1000 parts per million) that has a strong color, e.g., nickel or copper.

**Methylene blue**

<table>
<thead>
<tr>
<th></th>
<th>Time (sec)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Begin to Disappear</td>
</tr>
<tr>
<td><strong>Potting Soil</strong></td>
<td></td>
</tr>
<tr>
<td><strong>Sand</strong></td>
<td></td>
</tr>
</tbody>
</table>
### Chemical 1: ____________________

<table>
<thead>
<tr>
<th></th>
<th>Potting Soil</th>
<th>Sand</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Time (sec)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Begin to Disappear</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Disappear Totally</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### Chemical 2: ____________________

<table>
<thead>
<tr>
<th></th>
<th>Potting Soil</th>
<th>Sand</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Time (sec)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Begin to Disappear</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Disappear Totally</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Answer the following questions related to this experiment:

1. Which soil type absorbed the blue color faster? ________________________________

2. Why did you have to moisten the soil first?
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   _______________________________________________________________________
   _______________________________________________________________________

3. Develop a hypothesis to explain your results.
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   _______________________________________________________________________
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4. Relate your hypothesis to the concept of cation exchange capacity.
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   _______________________________________________________________________
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   _______________________________________________________________________
   _______________________________________________________________________

5. How do your results help to explain the changes in response by the earthworm to a chemical alone (Experiment 1) vs. in a soil (Experiment 3)?
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   _______________________________________________________________________
   _______________________________________________________________________
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6. Compare and contrast the observations for the methylene blue and the metal solution.
   _______________________________________________________________________
   _______________________________________________________________________
   _______________________________________________________________________
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   _______________________________________________________________________

7. If you tried more than one metal solution, is there a difference in the rate of disappearance between them? Suggest a reason for the difference.
   _______________________________________________________________________
   _______________________________________________________________________
   _______________________________________________________________________
   _______________________________________________________________________
S#12 – SENSORIMOTOR REACTIONS AND BEHAVIOR RESPONSES

Answer the following questions related to the experiments:

1. What does LOEC mean? In Experiment 1, at what pH did the worm react adversely? Develop an experiment that would allow you to calculate the LOEC for pH.

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2. How does the LOEC differ between just adding drops of chemical on the plastic raceway vs. adding the chemical into either potting soil or sand? Provide a hypothesis that could explain these differences.

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3. Compare the LOEC for each metal in potting soil vs. sand. If there is a difference, suggest an explanation. How might this idea be tested experimentally?

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4. Explain the concept of cation exchange capacity of soil. How does this change your hypothesis from question 3? Develop an experiment to test this new hypothesis.

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5. Describe the worm’s “decision-making process” when placed between the water and chemically treated sides.

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___________________________________________________________________________
6. Describe the worm’s reaction (toward or away) from each chemical in each of the three experiments.

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7. In Experiment 1, did the worm react similarly whether the worm moved forward or backward into the chemical? Develop a hypothesis to explain those observations that accounts for both the structure of the nervous system and ecological implications.

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8. Why does the data sheet want you to identify the LOEC as “between” rather than an exact value? What would you have to do to find the exact LOEC?

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9. What are the two types of acid rain? Develop an experiment that would test the hypothesis that the LOEC for both acid types is the same. Compare the LOEC of strong (e.g., HCl, H₂SO₄ or HNO₃) vs. weak acids (e.g., acetic acid [vinegar]).

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10. If a concentration is below the LOEC level, does that mean it is not toxic to the worm? Defend your answer by providing logical reasoning and scientific evidence to support your opinion. Design an experiment to test your hypothesis.

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___________________________________________________________________________
___________________________________________________________________________
Teacher Preparation for Class


2. Review the “Earthworm Video Guide” (C#5) and “Environmental Agents and Your Health” (S#13).

Segue

Briefly review earthworm structure and function and its importance for earthworm survival. Specifically, review normal and aldicarb affected neural-muscular interactions from “Nerve-Muscle Interactions and Sensory Neurons” (C#2).

Tasks To Be Completed During the Class Period

- Discuss the activities completed so far in the module and relate the structures and the work they do to the worm’s survival and the possibility that environmental agents affect the worm’s locomotion and its survival.
- Watch and discuss the Introduction and Part I – Preparing for the Experiment of the video/DVD using the “Earthworm Video Guide” (C#5) and relate to “Nerve-Muscle Interactions” (C#2).
- Set up the borrowing chamber test and examine the effects of ethanol on burrowing behavior.

Materials

WORKSHEETS

- “Earthworm Video Guide” (C#5)
- “Environmental Agents and Your Health” (S#13) - Assignment

MATERIALS FOR THE CLASS

- “Earthworm Burrowing: A Model for Chemical Effects on Locomotion” Video/DVD
- DVD player

Assignment

“Environmental Agents and Your Health” (S#13)
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

C#5 – Earthworm Video Guide

Teacher note: The information below provides the teacher with guidelines for instruction and information to supplement the video/DVD.

The effect of aldicarb on earthworm burrowing may be used as a model for the human health effects of aldicarb. While human nerve-muscle signaling is not biochemically identical to that of the worm, it is quite similar. Therefore, observation of aldicarb-exposed worms provides a life model for aldicarb’s human health implications. Because of its toxicity, aldicarb may not be used in classroom experiments. However the video provides a substitute experiment. It answers student questions and provides data for evaluating student hypotheses about the effect of different exposures of worms to aldicarb. The video with accompanying activities, therefore, becomes a “virtual inquiry experiment” and provides basic information about worms and environmental health. It models the effects a nerve poison (neurotoxicant) on locomotion mediated by nerve-muscle interaction. Aldicarb, a neurotoxicant used in this experiment, is in a class of chemicals called carbamates and is used as an insecticide to kill sucking insects. As you will see, it affects worm-burrowing movements. As you view this video, consider aldicarb a model of how chemicals in our environment can harm living organisms including humans.

The video begins with the burrowing of an untreated worm followed by the burrowing of aldicarb-treated worms. It asks the question, “Why the difference in burrowing?” The difference in burrowing (caused by aldicarb) is best understood in the context of normal nerve-to-muscle signaling. The video then proceeds to information about the experimental set-up for exposing worms to aldicarb and explains aldicarb-affected signaling. It concludes with segments designed to answer the student question, “What if a worm is exposed to higher/lower concentrations of aldicarb for a longer/shorter time?”

Because human movement is also mediated by nerve-muscle interaction similar to that of the earthworm, humans would likely experience difficulty moving just like earthworms. In fact, humans exposed to aldicarb have experienced locomotion problems similar to those of the worms seen in this video. Still used in food production, aldicarb is now subject to strict regulations. A reading included in this module “A is for Aldicarb, P is for Pesticide” (S#15) discusses aldicarb, its use, and effects. It is important to note that while users must follow regulations, the regulations must be enforced by governmental agencies subject to budget appropriations.

Three activities are integrated into this section: 1) a student activity, “How Does Aldicarb Work?” (S#16), that helps students analyze normal and aldicarb affected signaling; 2) a group “experimental” activity, “Burrowing Behavior of Aldicarb-Exposed Worms” (S#17); and 3) a classroom discussion activity, “What Might Happen If…?” (C#6), that summarizes student predictions about the effects of different exposures to aldicarb; that provides for student observations, conclusions, and analyses of the experimental trials shown in the video.
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

While watching Part II – Conducting the Experiment, discuss observations of the aldicarb-treated worms with varying exposures to concentration and time. The discussion will probably elicit this question: “What if the worm were exposed to a higher/lower concentration of aldicarb for a longer/shorter time.” Distribute “Burrowing Behavior of Aldicarb-Exposed Worms” S#17 before viewing portions of the video that address student questions. Students should record their observations including the time it takes for the worm to burrow out of sight. They should note any differences among exposed worms and between exposed and control worms.

DISCUSSION QUESTIONS:

1. Does treating the earthworm with aldicarb affect the time it takes for an impulse to travel down the worm/giant axons? (Answer: No, the impulse travel times were about the same.)

2. Is the speed of the impulse what determines the worm’s ability to escape predators? (Answer: If the impulse were slow it would affect the worm’s ability to escape. But the impulse speed is not the only thing that affects the worm’s ability to move.)

3. Did aldicarb affect the ability of the earthworm to move? Describe. (Answer: Aldicarb slowed down the worms and made their movements jerky. It also contorted their bodies.)
S#13 – ENVIRONMENTAL AGENTS AND YOUR HEALTH

Note to the teacher: Assign this reading as homework on Day 14. It will provide context for “A is for Aldicarb, P is for Pesticide” (S#15)

INTRODUCTION

Environmental agents include substances in our surroundings that may affect living organisms. We are generally unaware of many of them because they are present in our foods, drinking water or air in very small amounts.

Environmental agents may occur naturally in the environment or be made by humans and introduced into the environment. Living things exposed to these agents may change noticeably, change ever-so-slightly or not change at all. Scientists now understand that the kind and severity of change in an exposed organism is often determined by how old the organism was at the time of exposure, how long the organism was exposed, and by how much of the substance the organism was exposed to. Commonly, the earlier an organism is exposed, the greater the effect of the agent.

NATURALLY OCCURRING ENVIRONMENTAL AGENTS

Naturally occurring agents include table salt, nicotine and alcohol. Many natural substances are required for normal development. Among the examples above, salt is the only essential substance. But when present in the body in amounts too large or small, even an essential agent like salt may cause health problems for humans. For example, too much salt may result in high blood pressure.

Harmful health effects are caused not only by inappropriate amounts of essential agents, but also by nonessential ones like nicotine and alcohol. Nicotine in cigarette smoke and chewing tobacco causes addiction. In turn, addiction to smoking leads to heart disease, emphysema, cancer and stroke. Alcohol presents its own set of health problems. For instance, when exposed to alcohol, a baby in the womb is at risk of developing life-long abnormalities of its nervous and skeletal systems. As a consequence of these harmful effects, women who are pregnant or wish to become pregnant are advised not to smoke cigarettes or drink alcohol.

SYNTHETIC ENVIRONMENTAL AGENTS

Unlike natural environmental agents, “synthetic” environmental agents are substances that have been made or are being made in chemical laboratories and factories. Tens of thousands of these synthetic agents have been introduced into our surroundings through their production, use and disposal. Like natural agents, synthetic agents may or may not be harmful.

Our exposure to most synthetic agents is involuntary; we do not choose to be exposed to them, but are exposed because their production, use and disposal are not isolated from the general environment. Manufacturers and consumers - homeowners, farmers, governmental agencies and business owners - are responsible for their presence in our surroundings. Through their production and sale, manufacturers introduce these agents into our environment. Consumers release them when they use and dispose of them. Because consumers use and dispose of
S#13 – ENVIRONMENTAL AGENTS AND YOUR HEALTH

numerous synthetic substances – pesticides to kill insects, weeds or fungi; cleaning agents to remove dirt or kill bacteria or molds; and solvents to make, run or repair appliances, office equipment and production machinery - they are spread throughout our surroundings.

CONCLUSION

As a technical society, we have become dependent on synthetic environmental agents. However, many of these agents threaten our health and the health of our environment. The reading in this module, “A Is for Aldicarb, P Is for Pesticide,” focuses on one synthetic environmental agent of concern.
SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

CLASS DAYS 15 & 16 – VIDEO: OBSERVING THE EFFECT OF ALDICARB ON EARTHWORM BORROWING AND THE MEANING OF OBSERVED EARTHWORM BEHAVIORS FOR HUMAN HEALTH

Teacher Preparation for Class

1. Set up Video Part I for reference during discussion.
2. Review “Pesky Pests, Pesky Thoughts” (S#14), ”A is for Aldicarb, P is for Pesticide” (S#15), “How Does Aldicarb Work?” (S#16), “Burrowing Behavior of Aldicarb Exposed Earthworms” (S#17), and “What Might Happen If . . . ? ” (C#6).
3. Use “What Might Happen If…” (C#6) to help students reflect on the discussion and evaluate initial hypothesis.

Segue

Briefly discuss possible effects of environmental chemicals on crawling and burrowing of untreated earthworms and what behaviors students might expect to see from aldicarb treated earthworms.

Tasks To Be Completed During the Class Periods

- Finish and summarize observations from Day 12.
- Discuss reading “Environmental Agents and Your Health” (S#13).
- Do item “A” of “Pesky Pests, Pesky Thoughts?” (S#14).
- Read and discuss “A Is for Aldicarb, P Is for Pesticide” (S#15) and “How Does Aldicarb Work?” (S#16). You may wish to do this as a jigsaw reading activity.
- Finish and discuss “Pesky Pests, Pesky Thoughts?” (S#14).
- View Part III – Conducting the Experiment of the video/DVD and relate class observations to the aldicarb readings and to human health.
- Use the data sheet from “Burrowing Behavior of Aldicarb Exposed Earthworms” (S#17) to record qualitative observations and burrowing times. Begin graphs and answer questions and draw conclusions if time permits.
- Use student questions to lead the class in an inquiry discussion of “What Might Happen If . . . ?” (C#6).
## SECTION 2 — INSTRUCTIONAL GUIDE FOR TEACHERS

### Materials

**WORKSHEETS**

- “Earthworm Video Guide” (C#5)
- “Environmental Agents and Your Health” (S#13)
- “Pesky Pests, Pesky Thoughts?” (S#14)
- “A Is for Aldicarb, P Is for Pesticide” (S#15)
- “How Does Aldicarb Work?” (S#16)
- “Burrowing Behavior of Aldicarb Exposed Earthworms” (S#17)
- “What Might Happen If…?” (C#6)

**MATERIALS FOR THE CLASS**

- “Earthworm Burrowing: A Model for Chemical Effects on Locomotion” Video/DVD
- DVD player
A. How does aldicarb move from the soil into an insect? A human?

B. What does aldicarb do to the nervous system of an insect? A human?

C. What evidence is there that some of the aldicarb that is absorbed into the plant may eventually be eaten by humans?

D. What happens to the aldicarb that is placed in the soil but does not enter the plant?

E. How does aldicarb get into the water we drink?

F. Using the information from the answers to the questions above, explain how aldicarb might get into our bodies and how it may affect our health.
Note to the Teacher: Take a few minutes and have student groups read and discuss the Introduction and look at the Concept Map. As a class, briefly discuss student responses to the questions and Concept Map. Then assign each group one of the six sections of the reading, instructing them to complete the appropriate section of the concept map and prepare to report to the class what they learned (The more difficult sections of the reading may be assigned to two groups so checks on understanding come mostly from classmates.) Once groups are finished, have each group report to the class summarizing its section of the reading. Close the class discussion with a review of the completed Concept Map.

INTRODUCTION

In the reading that follows, you will learn about a chemical pesticide called aldicarb. The reading begins with basic information about pesticides. It then discusses how aldicarb is used, how it works, its regulation, and why people are concerned about aldicarb — as well as other pesticides used on food crops. The conclusion discusses how aldicarb has been regulated with the goal of preventing harm to humans.

Before you begin reading about aldicarb, think about and answer these questions:

1. How do insects react when you spray them with a pesticide like Raid™ or Spectracide™?
2. If sprays like Raid™ or other insecticides can kill insects in a matter of a few minutes, how strong or powerful are the chemicals used in the spray?
3. Could the health of a bird or other organism be affected if it were to eat many insects treated with a pesticide?

On the next page you will see a concept map covering the content of the reading on aldicarb. Some of the information has been filled in for you. As you read the article add information to the map in the spaces provided.
S#15 – “A IS FOR ALDICARB, P IS FOR PESTICIDE”

READINGS

1. Pesticides may be thought of as environmental agents. Environmental agents include physical agents like heat and light, chemical agents like salt and pesticides, and biological agents like bacteria, worms, elephants and molds. Our surroundings are full of environmental agents that might affect us and other living organisms — for good or ill.

2. Pesticides are chemical or biological agents used to kill pests like insects, mice, rats, and molds. Regardless of its target pest, the purpose of a pesticide is to kill the pest without harming the species being treated or “protected.” However, a pesticide may affect organisms other than those being treated or killed. Whether or not it affects other organisms depends on the make-up of the pesticide (whether it is designed to kill one kind of pest or many kinds of pests), how long the pesticide stays in the environment, and how it is transferred to the pest. Pesticides are delivered to pests in three main ways: sprayed directly on the pest (like many insecticides), presented in a food eaten by the pest (like ant traps or rat bait), or taken up by a living organism as it feeds on another living organism that has taken the pesticide into its body. Aldicarb is a pesticide of the last type. The pests eat plants that have taken in aldicarb as their roots take up water. Pesticides are among the most regulated environmental agents in the United States. Several federal governmental agencies regulate pesticides — their manufacture, distribution, use and environmental presence. The Federal agencies that regulate pesticides include the Environmental Protection Agency (EPA), the Food and Drug Administration (FDA), and the U.S. Department of Agriculture (USDA). These agencies are authorized by the U.S. Congress to enforce more than 14 separate regulations that govern pesticides. They are responsible for protecting public health and regulating materials that present possible hazards to humans or the environment. Aldicarb is one of many highly effective pesticides. Because it presents hazards to humans, it is regulated by several agencies.

3. Aldicarb Characteristics and Its Use on Plants
   Aldicarb is a chemical used around the world to kill plant pests that pierce and suck on plant leaves, stems, and roots. It contains five elements (carbon, hydrogen, oxygen, nitrogen and sulfur), is a white solid, and is extremely poisonous. It dissolves readily in water, but is not sprayed on crops because it could easily harm a person applying the pesticide. Instead, farm workers drill the highly toxic pesticide into the ground as a grainy powder. Because it is so toxic, by law only a trained, certified person may apply the pesticide. And only after a number of days have passed may a person enter a treated field.

   As the certified farm worker plants a row of seeds or treats a growing crop, a granular powder containing aldicarb is drilled about 5 to 7 centimeters into the soil near the seeds or plants. With the next rainfall, the aldicarb dissolves and plant roots take up the aldicarb-laced water. Once in the plant, the plant changes aldicarb into “metabolite” pesticides that are even more toxic than the aldicarb. As the plant sap moves throughout the plant, the aldicarb and its metabolites are distributed to all plant parts — roots, stems, leaves, blossoms and fruits. When small sucking organisms like mites, nematodes, and insects (aphids, whiteflies, leaf miners and others) pierce the plant tissue, they take in the plant sap containing the dissolved pesticides and are killed.
**S#15 – “A IS FOR ALDICARB, P IS FOR PESTICIDE”**

Because aldicarb and its metabolites stay in the plant for some time, feeding pests continue to die. As long as aldicarb and its metabolites are in the plant, the crop is protected from pests, and people who pick and eat the crop are at risk.

### 4. Government Regulation of Aldicarb Based on Scientific Studies

In an attempt to protect people from the harmful effects of aldicarb and its metabolites, governments regulate not only how aldicarb is applied to a crop, but also how long a grower must wait to harvest an aldicarb-treated food crop. In the United States, different food crops have different harvest wait times. For example, a grower must wait 30 days after treatment to harvest lemons, 120 days to harvest sugarcane, and 120 days to harvest sweet potatoes.

To set regulations like these, governmental officials rely on scientific studies. The studies include the best time to apply aldicarb to a particular crop, how much aldicarb can be used for that crop, how long aldicarb stays in different kinds of soil after being applied, and how long it takes a particular kind of plant to change aldicarb into harmless substances. As time passes, the amount of aldicarb (and its metabolites) in the plant will decrease. But some pesticide may remain at harvest time. Rules about application amounts, times and procedures are used in an attempt to protect people from eating food crops that might contain harmful amounts of aldicarb.

Based on scientific data, governments around the world make rules about the use of aldicarb. However, not all countries set the same rules and health protection standards because conditions are different around the world. And not all governments have the will and/or ability to check that all growers obey the rules. This means some crops treated with aldicarb may contain more aldicarb (and metabolites) than is allowed by government regulations. Food crops are not the only way people are exposed to aldicarb. It may also be in their drinking water. Runoff from rainfall carries some of the pesticide into rivers, streams and lakes. These surface waters are the source of drinking water for many people in the United States. In some areas of the nation, rainfall that soaks into the ground carries pesticide into groundwater that supplies the wells of many small towns and individual farms. Because aldicarb has contaminated some drinking water supplies, our national government has set a standard for how much aldicarb may be in our drinking water. The standard is now set at 0.003 mg of aldicarb/L (or 0.000003g/L) of drinking water. The goal for a future drinking water standard is 0.001 mg/L (or 0.000001g/L). But because the foods we eat may also be a source of aldicarb, the government has ruled that the total amount of aldicarb an adult may eat and drink in one day is 0.004 mg/kg body weight (or 0.000004g/kg). This standard means a 120 pound adult may eat and drink up to 0.220 mg (or 0.00022 grams) of aldicarb in one day.
5. Uses of Aldicarb and How It Works

Since 1970, aldicarb has been used to protect a variety of crops and help growers have plentiful harvests. Aldicarb is used mainly on cotton; other treated crops include bananas, grapes, grapefruit, hops, lemons, lettuce, oranges, peanuts, pecans, sorghum, soy beans, strawberries, sugar beets, sugarcane, sweet potatoes, tomatoes and decorative plants. Because too much of the pesticide was found in potatoes grown in the United States, its use on potato crops ended in the United States in 1987. However, because it works very well, aldicarb is still used to protect many crops from pests.

Aldicarb works well because it kills pests by upsetting the way their nerves signal muscles. Normally, nerves send an electrical message or impulse down the length of a nerve cell. When the impulse reaches the end of the nerve cell, a chemical is released at the end of the nerve cell. The chemical relays the nerve message across a small space to a muscle cell. In this way the muscle cell gets a signal to do its job, namely contract. To keep the muscle from being over-worked, a key enzyme destroys the chemical messenger. With the chemical messenger destroyed, the muscle cell relaxes and is able to receive the next message and do its job again. Aldicarb upsets this system. (See “Nerve-Muscle Interactions” C#2.)

Specifically, aldicarb keeps the enzyme from destroying the chemical messenger so the chemical messenger keeps on stimulating the muscle cell. This causes the muscle to repeatedly contract until it gets so tired it will not work. To feel what it is like to contract a muscle, make a tight fist — hold it for a few minutes. How do the muscles in your hand feel? If the particular muscles involved are used for breathing, those muscles will stay contracted and breathing will stop. The organism will die from a lack of oxygen. Aldicarb is a most effective pesticide because it affects other insect nerve pathways in the same way it affects the breathing pathway. All nerve pathways of a sucking insect looking for food are disrupted. The insect dies and is unable to reproduce. Aldicarb does an excellent job of killing pests, therefore disrupting nerve-muscle signaling in a host of species.

6. Aldicarb Use on Food Crops: A Reason for Concern

Aldicarb is of concern to people because in small amounts it acts on our nervous systems in the same way as it acts on those of insects. Aldicarb upsets our nerve messages by keeping the key enzyme from breaking down the chemical messenger. A person exposed to a very high amount of aldicarb may die or may arrive at an emergency room with a very slow heart rate or in a coma. With proper treatment, emergency room patients usually survive. People who have been exposed to aldicarb at a level higher than permitted (total daily intake of 0.004mg/kg of body weight) may experience dizziness, nausea, diarrhea, sweating, blurred vision, tiny pupils, difficulty breathing, shaky muscles and leg weakness. Usually these exposed individuals pass most of the aldicarb out of their body within 24 hours. However, the effects of long term exposure to low levels of aldicarb are unclear. Aldicarb has not been tested enough to state whether or not it or its “metabolites” might cause diseases of the nervous system or cancer due to eating contaminated food or drinking contaminated water.
In scientific experiments, earthworms exposed to amounts present in treated soil grow more slowly and have less body protein. Frogs exposed to a pesticide mixture including aldicarb showed a lowered immune response to a frog parasite. Women regularly exposed to aldicarb showed abnormal populations of a type of white blood cell (T-cell) that protects from body invaders. Very low doses of aldicarb given to pregnant rats affected the fetuses more than the mother; and pregnant rats kept the aldicarb in their bodies longer than did rats that were not pregnant. Yet three generations of rats exposed to higher doses showed no negative effects. Finally, in human cell cultures, aldicarb caused mutations and various kinds of damage to chromosomes.

7. Aldicarb in Wisconsin

In the 1970s and early 1980s, aldicarb was regularly used on potato crops in Wisconsin. In 1978, toxic wastes came to the surface of the ground in residential neighborhoods at Love Canal, New York. The event prompted people around the U.S. to check groundwater for contaminants. Studies soon documented many wells were contaminated with agricultural chemicals or materials that had moved out of landfills and dumps into the groundwater. In Wisconsin, Dr. Byron Shaw of the University of Wisconsin – Stevens Point discovered aldicarb in Wisconsin wells. His report prompted further testing of wells in areas where aldicarb was used. When aldicarb was found in more wells, the State Legislature formed a committee led by State Rep. Mary Lou Munts and a bill was drafted. By May, 1984, Wisconsin Governor Tony Earl signed the state’s groundwater protection law. It assured that all groundwater in Wisconsin would be protected. Rules for using aldicarb (ATC 30.24) were established in Wisconsin; these rules limit how much aldicarb can be used and require growers to inform the Wisconsin Department of Agriculture, Trade and Consumer Protection before they use it. Today, no one in Wisconsin is using aldicarb even though it could be used on crops like soybeans. Anyone using aldicarb in Wisconsin must obey the state rules that limit its use to 2 pounds of aldicarb per acre per year – every other year on a particular field. Wisconsin’s strict rules are meant to protect our food and drinking water from contamination, but not all our food and water comes from Wisconsin. We must therefore rely on national and international rules to protect us from harm caused by specific environmental agents.
S#16 – HOW DOES ALDICARB WORK?

Note to the Teacher: This exercise will help student pairs or groups analyze both normal and aldicarb-affected nerve-to-muscle signaling. Orient students to the illustrations below.

1. Using the key below, have students enter the name of each structure described in the first column (orange) in the table below.
2. Have students discuss and record the changes in appearance and position of each structure as one moves from a to b to c in the middle row of C#2.
3. Follow with a class discussion that clarifies what the changes mean for normal nerve-to-muscle signaling. Using this approach, have students analyze the effect of aldicarb on nerve-to-muscle signaling.

This diagram illustrates the sequence of events at a nerve-muscle synapse affected by the environmental agent aldicarb.

a) Chemical messenger (green half-capsules) is released at the end of the nerve axon (light brown) into the synapse (gap) between the nerve ending and the receiving, relaxed muscle (pink showing less overlap between filaments) with receptor sites (yellow).

b) Chemical messenger reacts with the receptor sites and stimulates the muscle to contract (overlap between filaments increases and muscle contracts).

c) The enzyme associated with the synapse (light blue arcs on the muscle) is inhibited by the environmental agent (dark blue arcs over light blue arcs) preventing the break-down of the chemical messenger and the muscle stays contracted (overlap between filaments stays increased). Contracted muscles cannot do the job for which they were intended; namely, contract to move a body part – engaged in lifting, walking, moving the eyes in their sockets, moving food through the gut, or breathing. Aldicarb’s effectiveness is a result of its ability to upset normal nerve-muscle signaling at the synapse, resulting in the inability of muscles to do this work. Aldicarb causes the death of insects that suck on plants as well as risk of illness or death in people who happen to ingest aldicarb.
S#16 – HOW DOES ALDICARB WORK?

STUDENT DIRECTIONS:

1. Using the key at the bottom of the illustrations about nerve-to-muscle signaling, enter the name of each structure listed in the first column below.
2. Look at the middle row of illustrations about normal signaling and beginning at “a” enter a description of each structure’s appearance and position.
3. Look at “b” and describe each structure’s appearance and position.
4. Finally, look, at “c” and describe the appearance and position of each structure.
5. With your group, review and discuss the changes from “a” to “b” and then “b” to “c.”
6. Repeat this process for aldicarb-affected nerve-to-muscle signaling as shown in the bottom row of illustrations of C#2.

NORMAL NERVE-TO-MUSCLE SIGNALING

<table>
<thead>
<tr>
<th>STRUCTURES</th>
<th>(A)</th>
<th>(B)</th>
<th>(C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>GREEN CAPSULE</td>
<td>Chemical messenger is released at end of nerve into the gap or synapse</td>
<td>Chemical messenger binds to/occupies receptor site on muscle</td>
<td>Chemical messenger is broken down by the key enzyme</td>
</tr>
<tr>
<td>Structure: Chemical Messenger</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>YELLOW OVAL</td>
<td>Site is free not bound/occupied by chemical messenger</td>
<td>Site is occupied by chemical messenger, message is conveyed</td>
<td>Site is free to receive more chemical messenger</td>
</tr>
<tr>
<td>Structure: Receptor Site on Muscle</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PINK/GREY STRINGS</td>
<td>Filaments separate, hardly meshed; muscle is relaxed</td>
<td>Filaments are interdigitated/highly meshed, muscle is contracted</td>
<td>Filaments separate/hardly meshed; muscle is relaxed</td>
</tr>
<tr>
<td>Structure: Muscle Filaments</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
ALDICARB-AFFECTED NERVE-TO-MUSCLE SIGNALING

<table>
<thead>
<tr>
<th>STRUCTURES</th>
<th>(A)</th>
<th>(B)</th>
<th>(C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>GREEN CAPSULE</td>
<td>Chem. messenger is released at end of nerve into gap/synapse</td>
<td>Chem. Messenger binds to/occupies receptor site on muscle</td>
<td>Chem. Messenger stays intact, is not broken down</td>
</tr>
<tr>
<td>Structure: Chemical Messenger</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>YELLOW OVAL</td>
<td>Site is free/not occupied by chemical messenger</td>
<td>Site is occupied by chemical messenger; message is conveyed</td>
<td>Site is occupied by chemical messenger and unable to receive a new message. The original message is still sent</td>
</tr>
<tr>
<td>Structure: Receptor Site on Muscle</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PINK/GREY STRINGS</td>
<td>Filaments more separated, muscle is relaxed</td>
<td>Filaments are interdigitated, muscle is contracted</td>
<td>Filaments remain highly meshed/interdigitated, message continues, muscle stays contracted and fatigues</td>
</tr>
<tr>
<td>Structure: Muscle Filaments</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

What structure in the illustrations represents the aldicarb? Where is it found? What does aldicarb do?

Key: The aldicarb is represented by a blue arc. It is located on the receptor site. It keeps the enzyme that breaks down the chemical messenger, from breaking down the chemical messenger so the muscle stays contracted and gets tired.
**S#17 – BURROWING BEHAVIOR OF ALDICARB-EXPOSED EARTHWORMS**

*Note to the Teacher:* The video showing aldicarb’s effect on earthworm burrowing also illustrates nerve-to-muscle signaling. (Exercise S#16 relates to normal and aldicarb-affected signaling and is provided to facilitate student understanding of the affected behaviors observed in the video.).

**PROBLEM:**
Does Aldicarb Affect Earthworm Burrowing Time?

**HYPOTHESIS:**

**EXPERIMENT:**
This experiment has been done in an approved laboratory; laws will not allow aldicarb to be used in classrooms.

**OBSERVATIONS/RESULTS:**

**TABLE OF BURROWING TIMES FOR ALDICARB-TREATED EARTHWORMS**

As you watch the earthworm burrow, enter in the appropriate box the length of time (in minutes and seconds) it takes the worm to burrow out of sight.

<table>
<thead>
<tr>
<th>TOTAL TIME TO BURROW</th>
<th>CONTROL – 0 PPM</th>
</tr>
</thead>
<tbody>
<tr>
<td>EXPOSURE TIME</td>
<td></td>
</tr>
<tr>
<td>2.5 MIN</td>
<td></td>
</tr>
<tr>
<td>5 MIN</td>
<td></td>
</tr>
<tr>
<td>10 MIN</td>
<td></td>
</tr>
<tr>
<td>20 MIN</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>TOTAL TIME TO BURROW</th>
<th>1.0 PPM</th>
</tr>
</thead>
<tbody>
<tr>
<td>EXPOSURE TIME</td>
<td></td>
</tr>
<tr>
<td>2.5 MIN</td>
<td></td>
</tr>
<tr>
<td>5 MIN</td>
<td></td>
</tr>
<tr>
<td>10 MIN</td>
<td></td>
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<tr>
<td>20 MIN</td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>TOTAL TIME TO BURROW</th>
<th>5.0 PPM</th>
</tr>
</thead>
<tbody>
<tr>
<td>EXPOSURE TIME</td>
<td></td>
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<tr>
<td>2.5 MIN</td>
<td></td>
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<td>5 MIN</td>
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<tr>
<td>10 MIN</td>
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<tr>
<td>20 MIN</td>
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</table>

<table>
<thead>
<tr>
<th>TOTAL TIME TO BURROW</th>
<th>10.0 PPM</th>
</tr>
</thead>
<tbody>
<tr>
<td>EXPOSURE TIME</td>
<td></td>
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<tr>
<td>2.5 MIN</td>
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<tr>
<td>5 MIN</td>
<td></td>
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<tr>
<td>10 MIN</td>
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<tr>
<td>20 MIN</td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>TOTAL TIME TO BURROW</th>
<th>20.0 PPM</th>
</tr>
</thead>
<tbody>
<tr>
<td>EXPOSURE TIME</td>
<td></td>
</tr>
<tr>
<td>2.5 MIN</td>
<td></td>
</tr>
<tr>
<td>5 MIN</td>
<td></td>
</tr>
<tr>
<td>10 MIN</td>
<td></td>
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<tr>
<td>20 MIN</td>
<td></td>
</tr>
</tbody>
</table>
S#17 – BURROWING BEHAVIOR OF ALDICARB-EXPOSED EARTHWORMS

OBSERVATIONS OF BURROWING OF ALDICARB-TREATED EARTHWORMS

As you watch the earthworms burrow, enter their burrowing times and describe their movements in the space below.
DIRECTIONS:
Select a color for the selected exposure time(s). Color the appropriate circle(s) in the Key below. Make a narrow bar graph for the selected concentration of aldicarb using the colors for the select exposure time(s). Then, using the same color, draw a line connecting the top of the 10 minute bars. Repeat the process for the other selected exposure time(s). Then answer the questions on the next page.

COLOR KEY | EXPOSURE TIME
---|---
_circle_ | 0 Minutes
_circle_ | 2.5 Minutes
_circle_ | 5 Minutes
_circle_ | 10 Minutes
_circle_ | 20 Minutes

KEY: Color the circle for each exposure time
S#17 – BURROWING BEHAVIOR OF ALDICARB-EXPOSED EARTHWORMS

QUESTIONS: EFFECT OF ALDICARB ON EARTHWORM BURROWING TIME

1. Does exposure to aldicarb affect the time it takes an earthworm to burrow? Explain your answer by referring to the experimental results.

2. Is there any concentration and exposure time at which you saw no effect of aldicarb? Explain your answer by referring to the experimental results.

3. Do you think the results of this experiment justify scientists’ concerns about using aldicarb on food crops? Explain your answer based on the experiment and the reading about aldicarb (S#7).
C#6 – WHAT MIGHT HAPPEN IF...?

Note to the Teacher: Show video/DVD. After viewing the control worms vs. time of exposure to dechlorinated tap water, have the class tell what they think about what they saw. Using 10 ppm aldicarb for 10 minutes exposure time as a reference, ask students what has been learned about how carbamate insecticides work on nerves and muscles, and engage them in a brainstorming activity that focuses on what might happen if a worm is exposed to different amounts of aldicarb for different lengths of time. Brainstorming provides students with the opportunity to articulate their own general question which may resemble the following:

“What might happen if a worm is exposed to ____________________________
(amount of aldicarb for ____________________________) period of time?”

Once students have determined the question they want to answer, it should be recorded and referred to as they hypothesize and discuss possible outcomes. You may want to use the chart below to summarize their thoughts before students view the relevant sections of Part III of the video/DVD showing the effects of different exposures to aldicarb.

This exercise renders student observations of the burrowing behavior an inquiry exercise. (The different doses and times of exposure also provide a model for student-planned investigations.)

<table>
<thead>
<tr>
<th>AMOUNT OF ALDICARB</th>
<th>0 PPM</th>
<th>___ PPM</th>
<th>___ PPM</th>
</tr>
</thead>
<tbody>
<tr>
<td>EXPOSURE TIME</td>
<td>STATE HYPOTHESIS AS COMPARISON BETWEEN CONTROL FOR SAME EXPOSURE TIME VS. SAME CONCENTRATION FOR DIFFERENT EXPOSURE TIME</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5 MIN</td>
<td></td>
<td></td>
<td></td>
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<td>10 MIN</td>
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<td></td>
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<td>20 MIN</td>
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</tbody>
</table>
INSTRUCTIONAL GUIDE PART III – OPTIONAL

STUDENT-DESIGNED EXPERIMENTS

Teacher Preparation for Class

1. Review “Guidelines for Planning Your Experiment” (S#18), “Can Earthworms Learn?” (S#19), and “What’s in a Word?” (S#20).
2. Be sure to have available the list of student-suggested, district-approved chemicals that may be tested for their effect on earthworms.

Segue

Remind students of the purpose of this student-planned activity, namely, students are to design and plan a test of an environmental chemical, such as ethanol, for its effect on earthworms and relate that to possible similar effects in humans.

Note: A set of experiments, “Can Earthworms Learn?” (S#19), may be done and used as the basis for student-designed experiments related to earthworm learning in addition to those about earthworm locomotion. Although this option extends the time frame for the module 2–3 days, it increases student interest.

Tasks To Be Completed During the remaining Class Periods

- Have students choose the one or two environmental agent(s) to be tested for their effect on worm locomotion (from among those approved by the school’s safety officer) OR begin “Can Earthworms Learn?” (S#19), OR complete “What’s in a Word?” (S#20) activity.
- Begin planning experiments on behaviors of earthworms treated with environmental chemicals using “Guidelines for Planning Your Experiment” (S#18).
- Finish planning experiments using “Guidelines for Planning Your Experiment”
- Organize equipment and prepare solutions for student planned experiments.
- Carry out student-planned experiments and bring them to closure.
EXAMINING THE EFFECT OF ETHANOL ON EARTHWORM BURROWING BEHAVIOR

Materials needed to test any effect aldicarb might have on the earthworm burrowing is listed here. Since the aldicarb experiment is not actually conducted in the classroom, additional materials are listed for an in-class experiment using the same protocol but for ethanol or other various liquid pesticides.

1 – 500 mL graduated cylinder
1 – 10 mL graduated cylinder
2 – Stop watches
Moist potting soil
2 – Plastic cups per student (12-16 oz)
Balance
4 worms per student
Aprons (1 per student)
Goggles (1 per student)
Nitrile gloves (1 pair per student)
Filter paper (5cm size, to match cup)
Paper for weighing
Dechlorinated tap water (1 gallon)
1 bottle 95% ethanol
4 – Glass bottles for serial dilutions to 1, 0.1, 0.01, and 0% ethanol
Pipets (graduated; if not available use medicine droppers and 10 mL graduated cylinder)
Spray bottles/cans of various liquid pesticides (e.g., Round Up, ant/roach killer)
S#18 – GUIDELINES FOR PLANNING YOUR EXPERIMENT

Note to the Teacher: Solutions for the student-designed experiments should be percent (%) solutions rather than molar solutions. See Solution Preparation Instructions, p. 236.

AS YOU PLAN YOUR EXPERIMENT REMEMBER TO INCLUDE THESE PARTS OF AN INVESTIGATION:

a) Problem Question  
b) Hypothesis  
c) Experiment  
d) Results/Observations  
e) Analysis of Results  
f) Conclusion

An analysis of the results often comes before the conclusion. The analysis can be the most interesting part of a report since it discusses different possibilities for explaining results (including scientific errors) and often leads to new questions and hypotheses. Below are details that will help you as you plan each part of your investigation. When your group believes it has done each item, check it and go on to the next item. (You probably will find the need to add to an earlier step as you work on a later step.) When you are done, if time permits, have another group check your plan. Finally, have your teacher check your plan and assign who will provide each piece of equipment.

PROBLEM QUESTION:

_____ 1. Clearly put into words the problem you are trying to solve. Do this in one sentence.  
_____ 2. Test only one factor in your experiment and be sure the problem question deals with that factor.  
_____ 3. Make sure you can work to solve the problem question over a period of two or three class days. Remember, well cared for worms will “work” while you are in other classes and at home!

HYPOTHESIS:

_____ 1. Write an answer to the problem question. It should be an educated guess based on your experiences and literature search.  
_____ 2. Express your hypothesis in the future voice, for example, “Earthworms treated with ______ for ___, ___, and ___, minutes will ______________.”  
_____ 3. Remember, your hypothesis does not change based on your results; once the experiment is done you will evaluate your hypothesis – tell whether it is correct or incorrect – based on your results. A hypothesis that proves untrue is acceptable!
S#18 – GUIDELINES FOR PLANNING YOUR EXPERIMENT

EXPERIMENT:

(Some activities you have already done contain these sections. Use them as models for your work.)

1. Write a Materials section that lists all the equipment and worms you will need.
2. Decide which equipment group members will provide and what you think your school will provide. Agree as to who will provide each item.
3. Write a numbered, step-by-step procedure that any person could follow if they wanted to repeat your experiment and check your results.

RESULTS/OBSERVATIONS:

1. Imagine all the things you are likely to observe. List them and plan space for recording them. You may want to make tables, graphs and leave space for drawings.
2. If you expect to get quantitative data (numbers) be sure your table provides for that data. (Hint: Use the table provided for the video on aldicarb-exposed earthworms in S#16 as an example.)
3. If you expect to make qualitative observations, provide space for writing what you might see.
4. Record your results so they are easy to locate and use.

ANALYSIS:

1. Write what you thought you would observe, why you thought you would make that observation, and whether you indeed did observe the expected.
2. If you did not observe what you expected to, tell what you did observe and why you think you observed the unexpected.
3. List any experimental errors you may have made and explain how they might have affected the results and how you might avoid them.
4. List any new questions one might investigate after doing this experiment.
5. Propose an hypothesis for each question you have listed.

CONCLUSION:

1. Write a statement that answers the problem question.
2. Base that statement on the results/observations of your experiment.
3. Evaluate your hypothesis – tell whether it was correct or incorrect, or whether you have enough information to evaluate your hypothesis!
S#19 – CAN EARTHWORMS LEARN?

BACKGROUND

If humans were unable to learn they would not be able to walk, talk and hold jobs – skills needed for survival. To learn these skills, we depend on a large brain, but with brains the size of a small pin head, you might wonder if the earthworm’s survival depends on its ability to learn. You have studied the earthworm’s crawling behavior and “startle response.” You will now test to see if earthworms can learn by “habituation,” one kind of learning typical of many organisms. In these experiments, habituation means the earthworm will not exhibit the “startle response.”

Every day humans habituate to sensory stimuli; for example, traffic noises. After hearing the noises from a busy street for a while, a person no longer hears the sounds of passing traffic – the individual has learned to ignore the sounds. In this experiment with the earthworm you will watch for a change in its response to a repeated sensory stimulus (like a touch) – even if the stimulus prompts the “startle response.”

In its natural soil environment, the worm would rapidly burrow into the soil to avoid touch or capture by a bird or fisherman. The next time someone is digging in your garden, try to catch a worm while it is in its native environment to convince yourself that the worm is a good escape artist. In this experiment, the worm is on a flat surface into which it cannot burrow, so you will be able to repeatedly touch the worm and provide it with the opportunity to learn.

Test I – PROBLEM: CAN AN EARTHWORM LEARN?

HYPOTHESIS (WRITE WHAT YOU THINK WILL HAPPEN):

__________________________________________________________________________

__________________________________________________________________________

__________________________________________________________________________

__________________________________________________________________________

__________________________________________________________________________

__________________________________________________________________________

__________________________________________________________________________

__________________________________________________________________________
S#19 – CAN EARTHWORMS LEARN?

Test I – PROBLEM: CAN AN EARTHWORM LEARN? (CONTINUED)

EXPERIMENT:

MATERIALS (PER GROUP FOR TESTS I, II, AND III)

- Water
- Clean cloth or sponge
- Waxed paper cup
- 3 earthworms at room temperature
- Moist paper towel
- Your activity sheet and pencil
- Dropper bottle of dechlorinated tap water (to keep worms moist)
- A clean, well-rinsed surface – a table, lab bench, or area of the floor
- Stopwatch or watch that displays tenths of a second

PROCEDURE:

Before beginning clean and rinse your work area well. Have all materials available and choose roles: Timer, Experimenter and Recorder. All will later record.

REMINDER: KEEP YOUR WORMS DAMP TO KEEP THEM FROM DRYING OUT AND SUFFOCATING.

1. Experimenter: Put one worm on the clean, well-rinsed surface.

2. Timer: Practice starting and stopping the stopwatch or reading seconds on a watch. When you are ready, tell the experimenter.

3. Experimenter: Stimulate the worm by touching its head. Do this by holding your hand about six inches away from the worm. Move your hand in to stimulate the worm with your thumb. Apply pressure like you would press the button of a remote control and then move your hand away. If the worm did not rapidly move away from your thumb, the timer should let you know when 15 seconds have gone by, then you should try again, using a slightly increased pressure. Once the animal gives a definite response, the timer should call out every 15 seconds and the experimenter should apply another stimulus.

4. Recorder: After the worm begins to respond, keep track of each stimulus on Table I. When the worm stops responding to three stimuli in a row, the animal has habituated to the stimulus – it has learned!

5. All: Enter the Recorder’s information in Table I, respond to questions 1 & 2, and write your “Conclusion” for this part of the activity.
S#19 – CAN EARTHWORMS LEARN?

Test I – PROBLEM: CAN AN EARTHWORM LEARN? (CONTINUED)

OBSERVATIONS/RESULTS I:

TABLE I

<table>
<thead>
<tr>
<th>STIMULUS</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
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<th>12</th>
</tr>
</thead>
<tbody>
<tr>
<td>“Startle Response” (Y/N)</td>
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</tbody>
</table>

1. Was the earthworm able to learn? ______ If so, what did it learn and what is the evidence that indicates it did learn?

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

2. Is the ability to learn important for earthworms? ______ Explain:

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

CONCLUSION I:

(Answer the problem question providing evidence from your activity and tell whether your hypothesis was correct or incorrect. Use complete sentences.)
Can an earthworm learn?________ Explain:

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________
S#19 – CAN EARTHWORMS LEARN?

Test II – PROBLEM: CAN AN EARTHWORM RELEARN A SKILL - FASTER THAN IT FIRST LEARNED THAT SKILL?

HYPOTHESIS (WRITE WHAT YOU THINK WILL HAPPEN):

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

EXPERIMENT:

MATERIALS (SEE TEST I MATERIALS)

PROCEDURE

1. Stop stimulating the worm for five minutes after it has habituated. Then apply the touch stimulus. (If the worm doesn’t respond, it has not forgotten what it learned; it is still habituated. Wait two more minutes and stimulate again. When the worm exhibits the “startle response,” it has forgotten what it learned and is no longer habituated. Can the worm relearn more quickly than it first learned?

2. Switch roles. (See Procedure, Test I.)

3. Repeat Steps 3 and 4 in Test I.

4. All: Enter the information in Table II, respond to items 1-5, and write your “Conclusion” for this part of the activity.

OBSERVATIONS/RESULTS II:

TABLE II

<table>
<thead>
<tr>
<th>STIMULUS</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
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<th>10</th>
<th>11</th>
<th>12</th>
</tr>
</thead>
<tbody>
<tr>
<td>“Startle Response” (Y/N)</td>
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</tbody>
</table>

1. Can an earthworm forget a learned behavior? _____ Provide evidence for your answer:

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________
S#19 – CAN EARTHWORMS LEARN?

Test II – CAN AN EARTHWORM RELEARN A SKILL - FASTER THAN IT FIRST LEARNED THAT SKILL? (CONTINUED)

2. Can an earthworm relearn a behavior? Provide evidence for your answer:

________________________________________________________________________

________________________________________________________________________

3. Check Tables I and II for the following information and record it below. Learning took _____ stimuli in Test I. Relearning took ______ stimuli in Test II. Compare and explain your answers.

________________________________________________________________________

________________________________________________________________________

4. Would habituation always be good for an earthworm? Explain your answer with an example.

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

CONCLUSION II:

(Answer the problem question providing evidence from your activity and tell whether your hypothesis was correct or incorrect. Use complete sentences.)

1. Can an earthworm relearn a skill and relearn it faster than it first learned that skill? Explain:

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________
S#19 – CAN EARTHWORMS LEARN?

TEST III – IS AN EARTHWORM HABITUATED TO A TOUCH ON ITS HEAD ALSO HABITUATED TO A TOUCH TO ITS TAIL?

HYPOTHESIS (WRITE WHAT YOU THINK WILL HAPPEN):

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

EXPERIMENT:

MATERIALS (SEE TEST 1)

PROCEDURE

1. Switch roles. (See Procedure, Test I)

2. Repeat Steps 3 and 4 in Test I, but this time touch the worm’s tail, not its head.

3. All: Enter the Recorder’s information in Table III, respond to the question below and write your “Conclusion” for this part of the activity.

OBSERVATIONS/RESULTS III:

<table>
<thead>
<tr>
<th>TABLE III</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>STIMULUS</strong></td>
</tr>
<tr>
<td>“Startle Response” (Y/N)</td>
</tr>
</tbody>
</table>

1. How did the habituated earthworm respond to a touch to its tail? Do your results demonstrate that the earlier learning involved the worm’s entire body?

________________________________________________________________________

________________________________________________________________________

2. Would this be helpful to the earthworm? Explain:

________________________________________________________________________
CONCLUSION III:

(Answer the problem providing evidence from your activity and tell whether your hypothesis was correct or incorrect. Use complete sentences.)
Is an earthworm that is habituated to a touch on its head also habituated to a touch to its tail? Explain:

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

ANALYSIS:

1. What does this set of activities demonstrate about the earthworm’s ability to respond to stimuli in its environment?

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

2. How is its ability to sense stimuli in the environment important to the earthworm?

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________
S#19 – CAN EARTHWORMS LEARN?

EXTENSIONS

A. Do you think the worm would remember as quickly if you waited a longer time (one hour or one day) before applying a stimulus the second, third, etc., time? If you want to try this experiment, put the habituated worm in a separate marked container with soil and set it aside for an hour or a day. Then repeat Test I above.

B. Test IV below.

TEST IV - IS AN EARTHWORM THAT HAS BEEN HABITUATED TO ONE KIND OF STIMULUS (TOUCH) ALSO HABITUATED TO A DIFFERENT KIND OF STIMULUS? (HEAT, CHEMICALS, LIGHT)

HYPOTHESIS (WRITE WHAT YOU THINK WILL HAPPEN):

________________________________________________________________________

________________________________________________________________________

EXPERIMENT:

MATERIALS

_________ Water
_________ Clean cloth or sponge
_________ Waxed paper cup
_________ 3 earthworms at room temperature
_________ Moist paper towel
_________ Your activity sheet and pencil
_________ Dropper bottle of dechlorinated tap water (to keep worms moist)
_________ A clean, well-rinsed surface – a table, lab bench, or area of the floor
_________ Dropper bottle of 7% ethyl alcohol (for test “a” below)
_________ Alcohol lamp and glass rod (for test “b1” below)
_________ Flashlight with intense narrow beam [maglight™] (for test “b2” below)
**S#19 – CAN EARTHWORMS LEARN?**

**TEST IV - IS AN EARTHWORM HABITUATED TO ONE STIMULUS (TOUCH) ALSO HABITUATED TO A DIFFERENT KIND OF STIMULUS? (CONTINUED)**

**PROCEDURE**

1. Habituate a worm to a touch on its head by repeating Test I.

2. Test the worm only one time using one of the following to answer the problem question:
   
   A. Chemical stimulus using ethyl alcohol: Using a dropper bottle of 7% ethyl alcohol, put a single drop of alcohol on the worm’s head. Did this stimulate a “startle response”?
   
   B. Physical stimuli
      
      1. Heat: Heat a glass rod in a cool flame so it gets hot but does not melt. Bring the rod close to the head of the worm but do not touch the worm’s skin. Does this stimulate a “startle response”?
      
      2. Light: Suddenly shine an intense beam of light on the worm’s head. Does this stimulate a “startle response”?

3. Enter data on Table IV. Then respond to items 1 thru 3 in the Analysis section.

4. Write your “Conclusion” for this part of the activity.

**OBSERVATIONS/RESULTS IV:**

**TABLE IV**

<table>
<thead>
<tr>
<th>STIMULUS</th>
<th>ALCOHOL</th>
<th>HEAT</th>
<th>LIGHT</th>
</tr>
</thead>
<tbody>
<tr>
<td>“Startle Response” (Y/N)</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**CONCLUSION IV:**

Is an earthworm habituated to one stimulus (touch) also habituated to a different stimulus (chemical or physical?) Explain:
S#19 – CAN EARTHWORMS LEARN?

TEST IV - IS AN EARTHWORM HABITUATED TO ONE STIMULUS (TOUCH) HABITUATED TO A DIFFERENT KIND OF STIMULUS? (CONTINUED)

ANALYSIS:

1. If an earthworm learns that one stimulus (a touch on its head) is not harmful, has it learned other stimuli are not harmful? Explain using the results from Test IV.

2. What does the earthworm’s response to the chemical and physical stimuli tell you about its ability to detect different kinds of environmental stimuli?

3. How is sensitivity to different environmental stimuli important to the earthworm?
You are shopping with your family at a garden store to buy what is needed for your vegetable garden. You think you might want to buy an environmental agent to keep pests from eating your vegetables. You find two products, read their names and wonder whether you should make a purchase. One label reads “plant protection product,” the other “Pesticide.” What information will help you decide whether to buy either?

**DIRECTIONS:**

Have a member of your group read the information on the grid to the group. As a group, discuss and answer each question in the grid. (You may use a dictionary for help.) Write your thoughts in the spaces provided. When you have finished the grid, discuss questions 1–5 and answer each question in the space provided. Finally, individually, write a paragraph telling whether you would buy either product. Also tell what helped you make that decision.

<table>
<thead>
<tr>
<th>PRODUCT DESCRIPTION</th>
<th>“PLANT PROTECTION PRODUCT”</th>
<th>“PESTICIDE”</th>
</tr>
</thead>
<tbody>
<tr>
<td>What does this description mean?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Does the description tell how the product works?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>What does the description emphasize about the product?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>How do you feel about using these environmental agents in your home or neighborhood?</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
1. Does either product name tell the user to be careful about using the product? Explain.

2. Which product name might lead to better sales? Explain.

3. Is it possible that a product could be called both a plant protection and pesticide? Explain.

4. If you were interested in using a product and protecting yourself from any possible harm, which name would you prefer? Explain.
S#20 – WHAT’S IN A WORD?

5. Some products that once were called “pesticides” (a chemical used to kill insects, spiders, rodents and other pests) are now called “protection products.”) How do you feel about the name change?

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

Would you make a purchase? What information would help you decide? (Use the information you have already developed to help you explain your decision.) Provide three reasons for your decision.

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________
SECTION 3 — SUPPORT MATERIALS

NATIONAL SCIENCE STANDARDS

INSTRUCTIONAL ACTIVITIES – STUDENT COPY MASTERS

S#1  Earthworms!!!
S#2  Earthworm Structures and the Work They Do
S#3  Normal Earthworm Responses to Threatening Environmental Stimuli – The “Startle Response”
S#4  Environmental Factors and Their Effect on Earthworm Movement
S#5  Sensing and Responding
S#6  What Do You Think?
S#7  Experiment 1 – Simple Sensorimotor Reactions
S#8  Experiment 2 – Preference Avoidance Behavior
S#9  Experiment 3 – Effect of Soil Type on Behavior Responses
S#10 Experiment 4 – Effect of Soil Type on Behavior Responses
S#11 Observing Cation Exchange Capacity of Soils
S#12 Sensorimotor Reactions and Behavior Responses
S#13 Environmental Agents and Your Health
S#14 Pesky Pests, Pesky Thoughts
S#15 A is for Aldicarb, P is for Pesticide
S#16 How Does Aldicarb Work?
S#17 Burrowing Behavior of Aldicarb-Exposed Earthworms
S#18 Guidelines for Planning Your Experiment
S#19 Can Earthworms Learn?
S#20 What’s in a Word?

MATERIALS AND POSSIBLE SOURCES

SOLUTION PREPARATION INSTRUCTIONS
SECTION 3 — SUPPORT MATERIALS

NATIONAL SCIENCE STANDARDS

PART 1 — STANDARDS-BASED, INQUIRY-BASED SCIENCE TEACHING AND LEARNING

This module was designed as an inquiry-based model of science teaching and learning. Human inquiry is a way of learning about the world and understanding the unknown through questioning, observing, and reasoning. Western science is a highly structured form of human inquiry.

The teaching of science is most effective when it is based on a set of developmentally appropriate standards for the content knowledge, skills, and dispositions that students can be expected to know and apply. In 2013, the National Research Council released the Next Generation Science Standards, which were based on the Framework for K-12 Science Education. These standards provide guidance for quality science education in the classroom.

Standards-based teaching involves the use of these standards to design, select, and adapt science curriculum. This module, The Neuro-Muscular Basis of Earthworm Movements: Effects of Physical and Chemical Environmental Agents has been designed to meet specific standards for science learning in grades 9-12. It has also been designed according to the standards for science teaching established in the National Science Education Standards. At the heart of these standards is the understanding that inquiry-based teaching is essential to student learning in science.

The science teaching standards that are most relevant to the development and implementation of this module are presented in the following section. The full compendium of the Next Generation Science Standards is available at: http://www.nextgenscience.org/next-generation-science-standards
SECTION 3 — SUPPORT MATERIALS

NATIONAL SCIENCE STANDARDS

PART 2 — NATIONAL SCIENCE TEACHING STANDARDS

STANDARD A
Teachers of science plan an inquiry-based science program for their students.

STANDARD B
Teachers of science guide and facilitate learning by:
- Focusing and supporting inquiries while interacting with students
- Orchestrating discourse among students about scientific ideas
- Challenging students to accept and share responsibility for their own learning
- Recognizing and responding to student diversity
- Encouraging and modeling the skills of scientific inquiry, as well as the curiosity, openness to new ideas and data, and skepticism that characterize science

STANDARD C
Teachers of science engage in ongoing assessment of their teaching and of student learning. They:
- Use multiple methods of assessment and systematically gather data about student understanding and ability
- Analyze assessment data to guide teaching
- Guide students in self-assessment

STANDARD D
Teachers of science design and manage learning environments that provide students with the time, space, and resources needed for learning science. Teachers:
- Structure the time available so that students are able to engage in extended investigations
- Create a setting that is flexible and supportive of science inquiry
- Ensure a safe working environment
- Make available the tools, materials, media, and technological resources at hand to support student learning in science
- Identify and use resources outside of the school or textbook
SECTION 3 — SUPPORT MATERIALS

STANDARD E

Teachers of science develop communities of science learners that reflect the intellectual rigor of scientific inquiry and the attitudes and social values conducive to science learning.

PART 3 — NATIONAL SCIENCE EDUCATION CONTENT STANDARDS: 9-12


Next Generation Science Standards (NRC 2013)

<table>
<thead>
<tr>
<th>Life Science</th>
<th>Scientific and Engineering Practices</th>
</tr>
</thead>
<tbody>
<tr>
<td>- The cell</td>
<td>• Asking questions</td>
</tr>
<tr>
<td>- Molecular basis of heredity</td>
<td>• Developing and using models</td>
</tr>
<tr>
<td>- Interdependence of organisms</td>
<td>• Planning and carrying out investigations</td>
</tr>
<tr>
<td>- Behavior of organisms</td>
<td>• Analyzing and interpreting data</td>
</tr>
</tbody>
</table>

Disciplinary Core Ideas – Life Sciences

- From Molecules to Organisms: Structures and Processes
- Variation of Traits: Genetic variation can result from mutations caused by environmental factors
- Ecosystems: Interactions, Energy, and Dynamics

Crosscutting Concepts

- Patterns
- Cause and effect
- Systems and system models
- Structure and function

Science as Inquiry

- Abilities necessary to do scientific inquiry
- Understandings about scientific inquiry

The Nature of Science

- Scientific investigations use a variety of methods
- Scientific knowledge is based on empirical evidence
- Scientific knowledge is open to revision in light of new evidence
- Science models, laws, mechanisms, and theories explain natural phenomena
- Science is a way of knowing
- Science addresses questions about the natural and material world
- Science is a human endeavor

Science and Society

- Connections between science research and societal concerns and issues
- Linking (minority) communities and science: science-based societal issues
SECTION 3 — SUPPORT MATERIALS

Based on the National Science Education Standards (National Research Council, 1996)

UNIFYING CONCEPTS AND PROCESSES

- **Evidence, models, and explanation:** As students begin to understand and apply more science concepts and processes, their understanding of what constitutes evidence will deepen. They will learn to use evidence to construct explanatory models — and use models to generate and test hypotheses. Their explanations will become more sophisticated, reflecting a rich base of science knowledge, evidence of logic, higher levels of analysis, and greater tolerance for criticism and uncertainty.

- **Systems, order and organization:** Through this module, student will begin to think and analyze in terms of systems. This thinking includes the understanding that systems have regularities and irregularities, differing levels of organization, and both within-system and out-of-system interactions.

SCIENCE AS INQUIRY

Students will develop the abilities necessary to:

- Identify questions and concepts that guide scientific investigations
- Design and conduct a scientific investigation
- Use technology and mathematics to improve investigations and communications
- Formulate and revise scientific explanations and models using logic and evidence
- Recognize and analyze alternative explanations and models
- Communicate and defend a scientific argument

LIFE SCIENCES

Students will develop a scientific understanding of:

- Cells have particular structures that underlie their functions
- The interdependence of organisms
- The behavior of organisms
SECTION 3 — SUPPORT MATERIALS

SCIENCE IN PERSONAL AND SOCIAL PERSPECTIVES

Students will develop a scientific understanding of the relationships between the environment, human activity, and societal decisions as they relate to:

- Personal and community health
- Natural resources
- Environmental quality
- Natural and human-induced hazards
- Science and technology in local, national, and global challenges

HISTORY AND NATURE OF SCIENCE

Students will engage in scientific investigations that illustrate and develop understanding of:

- Science as a human endeavor
- Nature of scientific knowledge

PART 4 — LEARNING BENCHMARKS

Concepts and Principles: Students will understand that:

1. Similarities among organisms are found in internal anatomical features, which can be used to infer the degree of relatedness among organisms.
2. In any environment, the growth and survival of organisms depends on the physical conditions of that environment.
3. Environmental agents can affect the course of development, affecting the survival of individual organisms and entire species.
4. Development is the process that occurs once an egg is fertilized. This process is systematic and predictable, and involves the specialization of many different cells to form tissues, organs, and organ systems.
5. The developing embryo of any organism encounters many risks in the form of environmental agents.
6. Environmental agents include both naturally occurring and introduced agents such as the heavy metals mercury and lead used or produced by humans as they attempt to enhance agriculture, manufacturing, and human life.
7. Toxic environmental agents have effects that may show up immediately or may show up many years later.
8. Humans are also affected by environmental agents.
9. Our environment may contain toxic levels of environmental agents.
10. Good individual health requires monitoring the soil, air, and water and taking steps to keep them safe for all of us.
SECTION 3 — SUPPORT MATERIALS

Scientific Inquiry and Processes: Students will understand that . . .
1. Scientific inquiry has some rules for what makes sense. It requires care in the use of words such as if, and, not, or, all, and some.
2. Explanations are a general rule summarizing how something works.
3. A single example, or even several examples, are not enough evidence from which to create a general rule.
4. A single example can be enough to show that even a general rule is not true.
5. Models are often used to think about processes that are difficult to observe as they happen.
6. It is important in scientific inquiry to keep honest, clear, and accurate records.
7. Hypotheses are valuable, even if they turn out not to be true.
8. Different explanations can be derived from the same evidence, and it is not always possible to know which explanation is the most likely.

Scientific Inquiry and Processes: Students will be able to . . .
1. Use appropriate tools, such as calculators and computers, to capture, store, retrieve, and analyze data.
2. Use appropriate instruments to make direct measurements and observations.
3. Organize information in tables and graphs, and explain the relationships they represent.
4. Read and interpret simple tables and graphs.
5. Locate and retrieve information from print and electronic sources.
6. Frame logical arguments, based on evidence.
7. Show skepticism of explanations and arguments that are not based evidence.

Science as a Human Endeavor: Students will know that . . .
1. The scientific challenge is to judge whether evidence is significant, and it often takes many repeated studies to finally reach a conclusion.
2. Scientific knowledge is constantly being modified and expanded. New information often challenges existing theories while new theories may lead to different interpretation of existing evidence.
3. Scientists differ greatly in what they study and how they study it. They don’t use one single method, but do use evidence, logical reasoning, and imagination in framing hypotheses and making sense of evidence.
4. What people expect to observe often affects what they actually do observe. Strong beliefs about what should happen can prevent one from seeing what actually does happen. Carefully designed investigations and collaboration with other investigators with different perspectives helps to avoid this pitfall.
5. People representing different ethnicities, cultures, classes, and genders have made important contributions to science. All of these contributions have been made available to others for scrutiny and criticism.
“Nobody loves me. Everybody hates me. Guess I’ll go eat worms.” So the jingle goes. Even though the song is meant to humor us when we are feeling sorry for ourselves, it is important that we like both ourselves and earthworms. After all, earthworms are among our best friends!

Earthworms unknowingly help us as they burrow underground to create their homes. In creating their tunneled homes, they mix and enrich the soil and make passages for air, water, and other organisms. Scientists estimate that there are about 50,000 earthworms in each acre of moist soil, or slightly more than one earthworm for about every square foot of ground. A soil with lots of dead plant and animal matter may contain over 20 earthworms per square foot. Yet how many of us think about them and all the good things they do every time we take a step outdoors?

We probably do not think of them because we normally do not see them when we are outdoors. However, if we look carefully, we may see earthworms on the ground at night when temperatures are moderate, light level is very low and the air is moist. Otherwise, they hibernate underground to survive cold or dry weather, or move through the soil to eat. As they eat they create tunnels. To make tunnels, the worm’s muscular prostomium (first segment) wedges into the soil and eats it. The muscular pharynx (throat) pushes the soil and any mixed-in dead plant matter further into the worm’s digestive tract. When the food and soil pass through the organs of the digestive tract, they are mixed and broken down into nutrients. The nutrient mix becomes available to plant roots once the worm excretes its wastes as “castings.” If you look carefully, you may find some castings, small coiled knots, on the ground near the opening of a worm burrow. Whether above or below ground, castings contribute to soil structure and fertility, providing the conditions and nutrients so important for plant growth.

Important to plants and other organisms are the air and water that enter the soil through earthworm burrows. Air is important for maintaining healthy plant roots and beneficial soil bacteria. Air in the soil is also very important to earthworms because they breathe through their moist skins. If the soil becomes water-logged, earthworms must move out of their burrows onto the ground to avoid drowning. Yet soil must contain some water if the earthworms’ skins are to be kept moist for breathing and moving (locomotion).

An earthworm moves through soil or on the ground using two sets of muscles: long ones that run the length of its body and ring-shaped ones that circle each body segment. When the long
S#1 – EARTHWORMS!!

muscles of the worm contract, the body of the earthworm becomes shorter and thicker. When the worm’s ring-shaped muscles contract, its body becomes longer and thinner. When you watch an earthworm crawl, you will notice that parts of the body may be thick and other parts are thin. You may also notice that the thickness of a given section of the body changes from thick to thin, and back to thick! For example, when the worm moves forward, its front extends forward and gets thin. Then, as the rear of the body is pulled forward, the head end thickens and the portion behind gets thinner. As the thinning and thickening pass along the length of its body, you can observe the wave-like pattern so characteristic of earthworm locomotion.

Because earthworms do not move great distances, a person wanting to improve soil needs to provide better conditions for earthworm health and reproduction. One effective way to improve soil is to increase the amount of dead plant material available to worms. Dead leaves or plant material may be layered on the soil as mulch or worked into the soil. Mulch will help hold soil moisture and temperature at levels favorable to earthworm activity. Working plant material into the soil provides food for worms, although it temporarily disturbs them. Another way to improve conditions for earthworms is to keep soil at a neutral pH. As the earthworm population increases, it will help maintain this desirable pH. The worms improve not only their environment, but ours as well! As long as they are fed, natural worm populations increase.

Earthworm populations are maintained or increased by a most interesting form of reproduction. While each earthworm has both male and female reproductive organs, most earthworms are not self-fertile; that is, the eggs of one earthworm cannot be fertilized by the sperm of that same earthworm. To reproduce, earthworms must mate. When earthworms are ready to mate, the largest body segment, the clitellum, changes from pinkish to red-orange in color. Two earthworms meet head to head and move along one another so their clitella are somewhat separated. Mucus is then secreted from the clitellum of each worm and the sperm from each worm moves into the storage pouch of the other worm. The mucus dries and forms a capsule into which more fluid is secreted. The worm then backs out of its capsule. As it does so, first the eggs and then sperm are deposited into the capsule; finally fertilization occurs. When the worm has fully left the capsule, the capsule remains in the soil and the fertilized eggs develop into baby worms.

Several factors are known to affect earthworm populations. Those already mentioned are soil temperature and moisture, soil pH, and the amount of dead plant matter available for food. Research has demonstrated that plowed fields have fewer earthworms than do pastures. Scientists have shown that placing fertilizer and insecticides in the soil when planting a row crop decreases worm populations near the row. As might be expected, the effect of agricultural chemicals on earthworm populations is different for each chemical. Chemicals like the pesticide aldicarb are known to be highly toxic to earthworms. As you progress through the module, you will observe the effect of aldicarb on earthworms. Earthworms respond to the chemicals that are dissolved in the water that surrounds soil particles. They may be crawl towards or away from these chemicals. While some of these chemicals, such as copper, manganese or zinc, are important in small amounts, they become toxic at higher concentrations. In this module, you will also observe how earthworms respond to different concentrations of these metal salts, as well as changes in soil pH (a measure of acidity).
**NAME**  
**S#2A/2B – EARTHWORM STRUCTURES AND THE WORK THEY DO**

Even though the earthworm looks simple, it has a fairly complex set of body systems and is able to appropriately respond to many different stimuli. In this activity, we are going to see how its body structures enable it to move. We are going to examine worm anatomy and see that underlying the worm’s simple appearance are some surprisingly complex structures - structures extremely well-adapted to the animal’s soil environment. First you will examine the external structure of the worm and relate observed structures to the worm’s crawling behavior. Then you will study its internal structures and relate those to its crawling behavior. Finally, you will summarize what you have learned about earthworm structures and their importance to the earthworm’s crawling motion and survival.

Before beginning the activity, look at the grid below. Enter information you already know. As you do the activity and readings, add information to complete the grid.

**GRID: COMPARING ANIMAL MOVEMENT AND SUPPORT**

<table>
<thead>
<tr>
<th></th>
<th>MOVEMENT</th>
<th>SUPPORT</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>EARTHWORM</td>
<td>Moves by contracting:</td>
<td></td>
<td></td>
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<tr>
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<td></td>
<td></td>
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<tr>
<td></td>
<td>and uses: to grip soil.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CATERPILLAR</td>
<td></td>
<td>Is supported by:</td>
<td></td>
<td></td>
</tr>
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<td></td>
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<td></td>
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<tr>
<td>SNAKE</td>
<td>Moves by contracting:</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>and uses: to grip soil.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HUMAN</td>
<td>Moves by contracting:</td>
<td></td>
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<td></td>
<td>Is supported by:</td>
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</tbody>
</table>
S#2A/2B – EARTHWORM STRUCTURES AND THE WORK THEY DO

DIRECTIONS:

- Before beginning the activity, clean and rinse your work area well.
- Have all materials available.
- Group members should take turns reading directions aloud.
- ALL group members will do each step of the procedure and record.

REMINders:

- Do not touch the worms more than necessary.
- Keep your worms damp so they do not dry out and suffocate.

MATERIALS (FOR EACH STUDENT GROUP)

For External Anatomy

- Water
- Clean cloth or sponge
- Activity sheet
- Dry, stiff paper towel
- 1 or 2 live earthworms in moist paper toweling in a paper cup
- Dropper bottle of dechlorinated tap water to moisten worms
- Dissecting microscope with an illuminating light
- Magnifying glass with a strong flashlight

For Internal Anatomy

- Long, skinny balloons
- Compound microscope with objectives of 4X - 40X
- Prepared microscope slides of earthworm cross sections
**S#2A – PROCEDURE: EXTERNAL ANATOMY**

1. As you work through this procedure, label the diagram at the end of this section with the underlined names provided in the directions below.

2. Clean your work area well with fresh water and a clean cloth or sponge. Put the live earthworm on your work area surface and carefully examine it with the magnifying glass. The most obvious characteristic of the worm is that its body is made up of a series of similar looking ringed structures. Each ringed structure is called a segment and it is this characteristic that gives these worms their name. (We are also made of repeating segmented structures. In our case, our body segments are different, but you can still see them in the repeating bones of our spines and ribs. Segmentation is a common feature of the body plan of many animal groups.) In the worm there is one segment that is much longer and thicker than the others. It is called the clitellum and is involved in sexual reproduction of earthworms. Worms are hermaphrodites (they are both male and female), yet they generally mate with another worm.

3. Carefully watch the earthworm move; you can probably tell the color and shape differences between the head and tail ends of the worm. Carefully examine the last segment and first segment (prostomium) of the worm. What are the differences?

   ________________________________________________________________

   ________________________________________________________________

   What are the jobs of the head end of the worm? How might those jobs account for the shape differences you noted above?

   ________________________________________________________________

   ________________________________________________________________

4. Touch and look at the top (dorsal) of the worm, and the bottom (ventral) of the worm. Note three differences.

   A. ________________________________________________________________

   B. ________________________________________________________________

   C. ________________________________________________________________

   It is important for you to note these differences because you will use them later as you observe earthworm behavior.
5. Use your fingers and gently flip the worm upside down. What does it do?

Is its motion smooth or jerky?

Keep a picture of this motion in mind. You will use it in later activities. This shows us another behavior of the worm. It can flip itself over onto its underside/front. (Interestingly, this same type of behavior is a developmental milestone of human babies. They first develop enough strength to flip themselves over onto their backs, but it takes them longer to be able to flip from their backs onto their front sides.) The ability of the worm to perform this behavior shows us that at least the worm can tell its back from its front.

6. Put the worm on a stiff dry paper towel. When it starts moving, put your ear close to the worm and listen. What do you hear?

The earthworm maintains traction with the ground by means of many fine bristles (the setae) on its underside/front. Rub the worm’s underside with your finger from head to tail. Then rub from tail to head? Note any differences.

Does the roughness change when the worm changes shape? What might control this?

7. Let the worm crawl and watch the setae as they and the earthworm move. Use the magnifying glass and a bright light to observe the setae. How are they shaped?

How does the movement of these fine bristles help the worm in its environment?

In addition to providing supports, muscles work to extend the setae from the worm’s body and to pull them back again. How many setae are in each segment? To answer this, you may need to observe the setae with the dissecting microscope.

Draw and label a few setae on the diagram of the worm.
8. Place the worm in moist paper toweling in the container and return it to the appropriate place.

9. Enter the information you have learned so far in the grid at the beginning of this exercise and be sure you label the worm below.

FIGURE 2: EARTHWORM EXTERNAL ANATOMY
**S#2B – PROCEDURE: INTERNAL ANATOMY**

**BACKGROUND**

In studying the locomotion of the worm, it was evident that the worm moved in waves of muscular contractions that passed smoothly along its body. You observed waves of alternate lengthening and shortening of a portion of the worm’s body. The lengthening is caused by contraction of circular muscles that squeeze on the inside of the worm to make it thinner and longer. The shortening and thickening are caused by longitudinal muscles that pull the extended portion of the worm together. Before looking at those muscles under the microscope, think about how they cause movement. All muscles must be attached to a structure to cause movement. Human muscles work on bones to which they are attached; or in the case of intestines, the muscles work on the food in the gut at the same time they are held in place by connective tissues.

From handling worms, you know there are no bones in a worm. What do the worm’s muscles attach to and work on? They attach to and work on a water skeleton. The water skeleton is found between the muscles and the gut and runs the entire length of the animal. **Label the water skeleton in Figure 2.** Then follow the procedure below to understand how the muscles work as the worm moves.

1. Take a long balloon and fill it with water. Put your hands around the outside of the balloon and squeeze it. This is the same action that the circular muscles have on the fluid-filled “skeleton” of the worm. What two things happen to the balloon as you squeeze?

   A. 

   B. 

   What would happen to the body of the worm?

   If you let go of the balloon, what does it do?

   The worm’s body does not behave in this way. Instead, it must use its longitudinal muscles to return to its original shape. This flexible, water-filled “skeleton” of the worm actually makes it possible for the worm to crawl through the soil. Using the water skeleton to push against, the worm can move its head into small openings. It makes those openings larger by thickening its body. It then pulls forward into the enlarged opening. This is the worm’s perfect solution for moving through its environment.
FIGURE 2
EARTHWORM CROSS-SECTION:
LOCATE THE WATER SKELETON

\[\text{FIGURE 2}\]

2. Put the slide of the earthworm cross-section on the microscope stage and make sure the light is shining through the thin slice of worm. Begin examining the cross-section with the lowest power objective (usually 4X). You can probably see the entire cross-section of the worm. The first thing to notice is that the top and bottom of the worm are different. The **dorsal blood vessel** or heart is on the back (top) of the animal and the **nerve cord** is on its ventral (bottom).

Around the outside of the animal are two layers of muscles: the **outer circular muscles and the inner longitudinal muscles**. We know they do different jobs to move the earthworm. Describe any differences in their structure. Relate the process of squeezing the water balloon to each of these muscle layers.

3. Locate the **nerve cord** of the animal. It is fairly small and located on the opposite side of the animal from the heart or dorsal blood vessel. Once you have located it with a low power objective, use higher power objectives to examine it. The nerve cord is the communication system of the animal. It triggers the muscles to act and coordinates action among neighboring segments. The nerve cord also receives and analyzes sensory signals so the animal can respond appropriately.

Why do you think the nerve cord is on the ventral (bottom) side? Compare and contrast to where the nerve cord is in snakes.

4. Develop a testable hypothesis to explain any differences.
S#2B – PROCEDURE: INTERNAL ANATOMY

The most obvious structures in the worm’s nerve cord are the three large circular shapes near its top. These are extremely large nerve fibers called giant axons. Most nerve fibers are very small. The giant axons’ large size allows them to send electrical signals extremely rapidly – much more quickly than the smaller nerve fibers. In fact, they are the nerve fibers involved in the escape or “startle response” so very important for the worm’s survival. (Try to estimate the size of the giant axons. You can do this by measuring the width of the entire worm slice with a ruler directly from the slide and estimating how large the nerve cord might be and then the giant axons themselves.

5. Later in the module, you may choose to investigate whether an environmental chemical affects the life-saving “startle response” of the worm. If it does, either the giant axons or the muscles themselves will have been affected by the chemical.

6. Because two earthworms need to be lined up in a particular fashion to reproduce, what would you expect to happen to the earthworm population if a chemical affected their ability to line up?
S#3 – NORMAL EARTHWORM RESPONSES TO THREATENING STIMULI – THE “STARTLE RESPONSE”

As the earthworm crawls along, it responds to its environment. Like us, it depends on both its environment and its ability to respond to stimuli in its environment. Some things in a worm’s environment that are important to it are not necessarily important to us and vice versa. What is important to understand is that we and earthworms are interdependent, so what affects the worm will to some extent affect us.

In later activities you will learn about specific environmental chemicals and their effect on the earthworm’s ability to respond. This activity allows you to observe normal earthworm responses to several stimuli. Later, when you check out the possible effect of a chemical, you will already know what normal behavior looks like. You will then be able to compare behaviors of untreated and treated worms. Remember, the earthworm’s nerve-muscle interactions are similar to yours, so the earthworm is a model for understanding possible human health effects.

Below is a **Prediction / Reflection Log**. First read the questions in the “Tests” column. Then think about each test and write your expectation of how the earthworm will respond to that test in the “My Hypothesis” column. When you have finished each test, return to the log and complete the column, “My Observations of Earthworm Behavior.”

### Prediction / Reflection Log

<table>
<thead>
<tr>
<th>WHAT DO I ALREADY KNOW?</th>
<th>WHAT DO I NEED TO KNOW (HYPOTHESIS DEVELOPMENT)?</th>
<th>WHAT DO I DO TO LEARN WHAT I NEED TO KNOW (METHODS DEVELOPMENT)?</th>
<th>WHAT DO I NOW KNOW (OBSERVATIONS AND DATA COLLECTION)?</th>
<th>WHAT DO I NOW KNOW (DATA ANALYSIS AND INTERPRETATION)?</th>
<th>WHAT DO I NOW NEED TO KNOW (NEXT EXPERIMENTS)?</th>
</tr>
</thead>
<tbody>
<tr>
<td>WHERE DO WORMS LIVE?</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HOW DO WORMS KNOW WHAT IS BAD FOR THEM?</td>
<td>What will an earthworm do when light is shone in its path? Behind it? On top of it? Different color light?</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HOW DO WORMS KNOW IT IS HOT OUTSIDE?</td>
<td>What will an earthworm do when alcohol is placed on/near its head?</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Before beginning, clean and rinse your work area well. Have all materials available and choose roles: Worm Handler, Equipment Manager, and Recorder. All will keep records. Switch roles at the end of each section of the procedure.

**REMINDERS:**

- Do not touch the worms too much because they will stop responding (This is the simple kind of learning known as habituation.).
- Once you start this set of experiments, use one worm until it stops responding and then switch to another worm while the first one recovers.
- Keep your worms damp or they will dry out and suffocate.

**MATERIALS:**

- Container (paper cup) with three earthworms in moist paper toweling at room temperature
- Water
- Clean cloth or sponge
- Glass rod
- Dropper of dechlorinated tap water to keep animals moist
- Matches
- Small dropper bottle of 7% ethanol (ethyl alcohol)
- Narrow-beamed halogen desk lamp, Maglight™ or microscope illuminator for intense beam of light
- Alcohol lamp or a butane lighter for a cool flame
S#3 – NORMAL EARTHWORM RESPONSES TO THREATENING STIMULI

A. PROCEDURE: LIGHT STIMULUS

1. Read questions 1 through 6 before you proceed.

2. **Equipment Manager:** Clean and rinse your work area well.

3. **Worm Handler:** Put a worm on your work area and let it crawl. Make sure it is moist.

4. **Equipment Manager:** When the worm is moving along nicely, shine a narrow flashlight beam in the path ahead of the worm, *not on the worm.*

5. **All:** Carefully observe what the worm does when its head enters the beam of light and discuss the worm’s response as a group.

6. **Recorder:** Summarize the group’s observation, and complete item 1 in the “Observations” section. Let the worm rest for a minute.

7. **Equipment Manager:** Shine the beam *on the worm’s head.*

8. **All:** Carefully observe the worm’s response to the light shone on its head and discuss your observations.

9. **Recorder:** Summarize the group’s observation, and do item 2.

10. As a group, complete all items.

**OBSERVATIONS: RESPONSE TO LIGHT STIMULUS**

1. How does the earthworm respond when it’s head enters the light?

   ____________________________________________________________

2. How does the earthworm respond when the light beam is directed at the middle of its head?

   ____________________________________________________________

3. Think about where the worm lives and what it does to survive. Does its response to light make sense? Explain.

   ____________________________________________________________
   ____________________________________________________________
S#3 – NORMAL EARTHWORM RESPONSES TO THREATENING STIMULI

4. How is this response important for the worm’s survival?

5. If the worm had vision like ours, would that be helpful for its survival?

6. Can you think of reasons why vision like ours might actually be harmful to the worm’s chances to survive in its environment?

B. PROCEDURE: CHEMICAL STIMULUS

1. Read questions 1 through 3 before you proceed.
2. Worm handler: Put a worm on your work area and let it crawl. Make sure it is moist.
3. Equipment manager: When the worm is moving along nicely, put a single drop of 7% ethanol on the worm’s head.
4. All: Carefully observe what the worm does when the alcohol is put on its head and discuss what you observed.
5. Record what you observe and complete questions 1 - 3 in “Observations.”

OBSERVATIONS: RESPONSE TO ALCOHOL/CHEMICAL STIMULUS

1. How does the worm respond to the drop of alcohol placed on its head?

2. How is this behavior similar to its response to light shone on its head?
3. How is this behavior different from that to light shone on its head?

C. PROCEDURE: HEAT STIMULUS

1. Read questions 1 through 5 before you proceed.
2. Equipment Manager: Clean and rinse your work area well.
3. Equipment Manager: Light the alcohol lamp or butane lighter and warm the glass rod in the flame. Do not melt the glass.
4. Worm Handler: Put a worm on your work area and let it crawl. Make sure it is moist.
5. Equipment Manager: When the worm is moving along nicely, bring the rod close to the head of the worm but do not touch the worm’s skin.
6. All: Observe the worm’s response as a group. Discuss what you observe. Then record your discussion. Complete items in the “Observations” sections below.

OBSERVATIONS: RESPONSE TO HEAT

1. How does the worm respond to the warm rod placed near its head?

2. How is this behavior like the worm’s response to light shone on its head?

3. How is this behavior different from its response to light shone on its head?

4. How is this behavior like the worm’s response to ethanol placed on its head?
S#3 – NORMAL EARTHWORM RESPONSES TO THREATENING STIMULI

5. How is this behavior different from its response to ethanol?

__________________________________________________________________________

GENERAL OBSERVATIONS

1. How are these responses different from crawling?

__________________________________________________________________________

2. Complete the second column of the Prediction / Reflection Log found at the beginning of this activity. Then answer questions 1 – 3 below:

HOW WOULD YOU RESPOND TO THESE STIMULI?

1. Do you find some of these stimuli irritating or harmful?

__________________________________________________________________________

2. Describe how you would respond to one of these stimuli.

__________________________________________________________________________

3. Review your responses to “Environmental Factors and their Effect on Earthworm Movement” S#3b and tell if you have changed your mind about whether some environmental factors affect earthworm movement.

__________________________________________________________________________

REMEMBER:
COMPLETE THE LOG AT THE BEGINNING OF THIS ACTIVITY and write a general description of the earthworm’s response to the three stimuli below or on the back of this page.
S#4 – ENVIRONMENTAL FACTORS AND THEIR EFFECT ON EARTHWORM MOVEMENT

DIRECTIONS

Look at the information in the grid below. As you think about each factor, place a check in either the “yes,” “no” or “uncertain” column to indicate your expectation of whether each factor would affect the worm’s speed.

ENVIRONMENTAL FACTORS AND THEIR EFFECT ON EARTHWORM MOVEMENT

<table>
<thead>
<tr>
<th>FACTOR</th>
<th>WOULD THIS AFFECT WORM MOVEMENT?</th>
</tr>
</thead>
<tbody>
<tr>
<td>Would the movement of an earthworm be affected by…</td>
<td>YES</td>
</tr>
<tr>
<td>The temperature of the soil and air around the worm?</td>
<td></td>
</tr>
<tr>
<td>The amount of light in the worm’s environment?</td>
<td></td>
</tr>
<tr>
<td>The amount of moisture on the ground and in the soil?</td>
<td></td>
</tr>
<tr>
<td>Chemicals that might be in the soil and the worm’s food?</td>
<td></td>
</tr>
<tr>
<td>The location of the earthworm in the soil or the ground?</td>
<td></td>
</tr>
</tbody>
</table>

WITH A BRIEF STATEMENT, EXPLAIN TWO OF YOUR CHOICES.

1. __________________________________________________________________________

2. __________________________________________________________________________
Anticipation Questions:

1. What signals do you pick up from (stimuli do you detect in) your environment?
2. Which of these signals (stimuli) is beneficial? Harmful? How do you respond to them?
3. Is it likely you are unaware of some signals from or changes in your environment?
4. Are you able to respond to stimuli/changes you do not know about? How do you feel about that?

As the earthworm crawls along, it responds to its environment. The worm’s survival, like ours, depends on its ability to respond to what happens in its environment. While worms and humans are able to respond to many changes in the environment, neither is able to respond to all changes. Naturally, human and earthworm responses are limited to the stimuli each is able to sense. Some stimuli in a worm’s environment that are important to it are not necessarily important to us and vice versa. For example, an earthworm has special cells in its skin that detect vibrations in the soil made by its predators. Humans cannot detect these vibrations. Generally, each life form has a range or set of stimuli that it can naturally sense and respond to.

Usually stimuli that an organism can sense are important for its survival in a natural setting. These detectable stimuli may be threatening or non-threatening. The proper responses to them usually contribute to the survival of the organism. Harmful stimuli are usually avoided. But if an organism cannot sense a harmful change in the environment, it cannot respond. For that organism, it is as if there has been no change in the environment – unless the life form can use its intellect/intelligence to understand that a change has taken place. Only with knowledge can an organism respond to environmental changes it is unable to sense. If the change is beneficial to the organism, survival is not an issue. But if the change is potentially harmful, only the informed organism understands and has the opportunity to act to eliminate the undetectable harm or avoid its dangers. Unlike most other organisms, we humans are capable of informing ourselves. How do we deal with substances we cannot sense but know to be harmful?

Carbon monoxide serves as an example. It is a harmful substance we cannot sense; we are unable to see, smell or taste this gas, yet it can kill us because it interferes with delivery of oxygen to our cells. For centuries coal miners in particular have been aware they could die from working in underground mines. Without knowing what in particular was causing their symptoms of achy muscles, fatigue, headache, dizziness, disorientation, and even death, miners learned to use a more sensitive animal to warn them of possible harm. Specifically, they carried canaries or mice.
that were highly-sensitive to carbon monoxide into the coal mines to warn of danger. If the canary or mouse fainted, miners knew they were in great danger and needed to leave the mine.

Today, mines and other workplaces are outfitted with devices that detect carbon monoxide. In our homes we protect ourselves by properly installing and maintaining appliances that burn natural gas, oil or kerosene. Our public health and fire departments help protect us by telling us that if all the members of a family are headachy, fatigued, dizzy or disoriented, carbon monoxide poisoning might be the cause and the family should seek immediate medical help. By knowing the symptoms of carbon monoxide poisoning and seeking help when we experience them, we are able to protect ourselves from a harmful substance we cannot sense. Humans are fortunate to have the brainpower to protect themselves from harm they cannot detect with their senses. But most organisms have little ability to respond and protect themselves if they cannot sense a harmful change in their environment.

In the investigation that follows this reading, you will observe the worm’s response to harmful stimuli it senses. In later activities you will learn about specific environmental chemicals and their effect on the earthworm’s ability to respond. As you do these activities, think about the earthworm’s ability to protect itself from the chemicals to which it is exposed. Also think about your ability to detect and protect yourself from the chemicals to which you are exposed. Are you aware of what you are exposed to? If you are aware, is it because your senses detected and informed you or because of independent information provided to you? This is why warning labels and symbols like “Mr. Yuck” and the skull and crossbones are on products. What do you do about these exposures? Remember, the earthworm’s nerve-muscle interactions are similar to yours. What affects their nerve-muscle interactions may affect yours. The earthworm is a model for understanding the possible human health effects from exposure to environmental substances.
1. Now that you have tested the earthworm’s response to three different threatening stimuli, think about how important it is for the worm to be able to both sense and respond to these stimuli. Using what you have learned, write a short paragraph that answers the following questions: Why is it important for the earthworm to be able to sense and respond to stimuli? What would it mean for the earthworm if it were unable to detect or respond to a threatening stimulus?

2. Do you think there are stimuli in the earthworm’s environment that it is unable to detect or respond to? Explain.

3. Are there things in the earthworm’s environment that affect its ability to sense or respond to stimuli? Explain.

4. If there are substances in its environment that affect either the worm’s ability to sense or respond to a stimulus, what might that mean for the earthworm?

5. If there are substances in our environment that affect our ability to sense or respond to a stimulus, what might that mean for us?
S#7 – EXPERIMENT 1 – SIMPLE SENSORIMOTOR REACTIONS

PROCEDURE:

**Note:** After each worm is used for a trial, rinse it in clean, dechlorinated water in a squirt bottle and place it in a beaker of moist soil. By rinsing the worm’s surface of acid or metal solution, the worms can be reused for the next set of experiments, as well as for the next class.

**A. Effect of pH:** With a transfer pipet, create a circle of drops of pH 7 (dechlorinated water or distilled water) on the plastic raceway that is large enough to encircle your worm. **Why should you use dechlorinated water rather than water directly from your tap?** Place worm on the raceway within the circle of drops. On your data sheet record the worm’s reaction (Strong, Mild, or No Reaction) when it comes into contact with the solution. Repeat for a total of 4 worms. When finished, wash the plastic raceway and dry with a paper towel. Repeat this procedure for pH 9 and 4.

**Optional Experiments:**

1. Use additional solutions of pH 5, 6, 8 and 10 to create a more complete picture of the worms’ sensitivity to acids and bases.
2. Compare the reactions between a strong acid (HCl or H₂SO₄) vs. a weak acid (acetic acid/vinegar) at pH 4, 5 and 6.

**B. Effect of Metals:** Make all your serial dilutions of the each of the 4 metal solutions as described above. Label each test tube with the metal concentration, e.g., 1000 ppm Cu, 100 ppm Cu, 10 ppm Cu, etc. Starting with the LOWEST concentration first, repeat methods outlined in part A above for a total of 4 different worms/concentration. Rinse the raceway between each change in concentration. Record your observations on your data sheet. **Once the worm has reacted STRONGLY to the metal solution, you do not need to test higher concentrations.** Metals should be chosen such that all four metals are tested by the class but any one group does 1-2 metals.
**S#7 – EXPERIMENT 1 – SIMPLE SENSORIMOTOR REACTIONS DATA SHEET**

Date: _______________  Hour: _______________  Acid: _______________________

Observers: ________________________________________________________________

A. Effect of pH

<table>
<thead>
<tr>
<th>pH</th>
<th>Worm #</th>
<th>Strong Reaction</th>
<th>Mild Reaction</th>
<th>No Reaction</th>
</tr>
</thead>
<tbody>
<tr>
<td>4</td>
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<td>4</td>
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<tr>
<td>9</td>
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<td>4</td>
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</tr>
</tbody>
</table>

**LOEC =** Lowest Observed Effect Concentration

LOEC value for ____________ acid is between pH __________ and __________.

**OPTIONAL: Be an Environmental Toxicologist!**
- Develop and conduct an experiment to find a more exact LOEC value!
- Compare the effects due to a strong acid vs. weak acid.
S#7 – EXPERIMENT 1 – SIMPLE SENSORIMOTOR REACTIONS DATA SHEET

Date: _______________       Hour: ____________________

Observers: ____________________________________________________________________

B. Effect of (Name of First Metal Tested):__________________________________________

<table>
<thead>
<tr>
<th>Concentration (ppm)</th>
<th>Worm #</th>
<th>Strong Reaction</th>
<th>Mild Reaction</th>
<th>No Reaction</th>
</tr>
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<tbody>
<tr>
<td>1000</td>
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</tbody>
</table>

LOEC = Lowest Observed Effect Concentration

LOEC value for ___________ is between __________ and __________ parts per million (ppm)
S#7 – EXPERIMENT 1 – SIMPLE SENSORIMOTOR REACTIONS DATA SHEET

Date: _______________       Hour: ____________________

Observers: __________________________________________________________________

B. Effect of (Name of Second Metal Tested): ________________________________

<table>
<thead>
<tr>
<th>Concentration (ppm)</th>
<th>Worm #</th>
<th>Strong Reaction</th>
<th>Mild Reaction</th>
<th>No Reaction</th>
</tr>
</thead>
<tbody>
<tr>
<td>1000</td>
<td>1</td>
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</tbody>
</table>

LOEC = **Lowest Observed Effect Concentration**

LOEC value for ___________ is between __________ and __________ parts per million (ppm)
S#8 – EXPERIMENT 2 – PREFERENCE AVOIDANCE BEHAVIOR

EXPERIMENT 2A: PROCEDURE

In this experiment, the worm can choose between different pH solutions, and then different concentrations of a metal solution to determine which side it will prefer.

**Note:** After each worm is used for a trial, rinse it in clean, dechlorinated water in a squirt bottle and place it in a beaker of moist soil. By rinsing the worm’s surface of acid or metal solution, the worms can be reused for the next set of experiments, as well as for the next class.

**D. Effect of pH:** With a transfer pipet, create 2 lines of drops as in the figure below, one of pH 7 (dechlorinated water or distilled water) **AND** one of either pH 4 or 9 on the plastic raceway that is large enough to encircle your worm. **Why should you use dechlorinated water rather than water directly from your tap?**

Place worm on the raceway between the line of drops.

**E.** On your data sheet record the worm’s reaction (Strong, Mild, or No Reaction) when it comes into contact with the solution. Does it “prefer” one pH over the other? Repeat for a total of 4 worms. When finished, wash the plastic raceway and dry with a paper towel. Repeat this procedure for but use the other pH solution.

**F. Effect of Metals:** Make all your serial dilutions of the each of the 4 metal solutions as described above. Label each test tube with the metal concentration, e.g., 1000 ppm Cu, 100 ppm Cu, 10 ppm Cu, etc. Starting with the **HIGHEST** concentration first, repeat methods outlined in parts A and B above for a total of 4 worms/concentration. **Once the worms seem to be choosing either side at random, i.e., they don’t seem to be**
avoiding or preferring one side over the other, you do not need to test lower concentrations. Rinse the raceway between each change in concentration. Record your observations on your data sheet. Metals should be chosen such that all four metals are tested by the class but any one group does 1-2 metals.

EXPERIMENT 2B: PROCEDURE

In this experiment, the worm can choose between distilled water and a particular solution of a chemical. In other words, will the worm sense which side it will prefer.

PROCEDURE:

D. Place 1 pre-cut paper towel at each end of the raceway. With a transfer pipet, moisten (not soak) one towel with dechlorinated or distilled water (Figure Q). You can use the same worms from Experiment 1.

E. Effect of pH: With a transfer pipet, moisten (not soak) the paper towel on the other end of the raceway with pH 9. Place the worm on the bare plastic between the two towels. Record on your data sheet which side it chooses. Repeat for a total of 4 worms. Dispose of the towels into the trash, rinse and dry the raceway. Use same procedure for pH 4 and vinegar.

F. Effect of Metals: Make all your serial dilutions of the each of the 4 metal solutions as described above. Label each test tube with the metal concentration, e.g., 1000 ppm Cu, 100 ppm Cu, 10 ppm Cu, etc. Starting with the HIGHEST concentration first, repeat methods outlined in parts A and B above for a total of 4 worms/concentration. Once the worm has stopped reacting to the metal solution, you do not need to test lower concentrations. Rinse the raceway between each change in concentration. Record your observations on your data sheet. Metals should be chosen such that all four metals are tested by the class but any one group does 1-2 metals.
Figure Q: Diagrammatic View for Experiment 2 Methods
S#8 – EXPERIMENT 2 – PREFERENCE AVOIDANCE BEHAVIOR DATA SHEET

Date: _______________     Hour: ____________________    

Observers: ______________________________________________________

EXPERIMENT 2A: PROCEDURE

A. Effect of pH

For each row, place a check in the box labeled # Observations each time you observe a worm choosing a specific pH value among the choices given in that row. Enter the number of total check marks for that pH choice in the box labeled Total Observations. An example is provided in the row labeled Sample Recording of Data.

<table>
<thead>
<tr>
<th>pH Choice</th>
<th># Worms Found In:</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>pH 4</td>
<td>pH 7</td>
<td>pH 9</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td># Observations</td>
<td>Total Observations</td>
<td># Observations</td>
<td>Total Observations</td>
<td># Observations</td>
<td>Total Observations</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
<td>/</td>
<td>1</td>
<td>///</td>
<td>3</td>
<td></td>
<td></td>
</tr>
<tr>
<td>pH 4 vs. pH 7</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>X</td>
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<tr>
<td>pH 9 vs. pH 7</td>
<td></td>
<td></td>
<td></td>
<td>X</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
EXPERIMENT 2A: PROCEDURE (continued)

B. Effect of Metal Concentration

Name of Metal Tested: ________________________________

<table>
<thead>
<tr>
<th>Concentration (parts per million)</th>
<th># Worms Found in:</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control Side (Water)</td>
<td># Observations</td>
<td>Total</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
<td></td>
<td>/</td>
<td>1</td>
</tr>
<tr>
<td>1000</td>
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<tr>
<td>100</td>
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<td></td>
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<tr>
<td>0</td>
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</tr>
</tbody>
</table>
EXPERIMENT 2B: PROCEDURE

A. Effect of pH

Acid Used: ____________________________________

For each row, place a check in the box labeled # Observations each time you observe a worm choosing a specific pH value among the choices given in that row. Enter the number of total check marks for that pH choice in the box labeled Total Observations. An example is provided in the row labeled Sample Recording of Data.

<table>
<thead>
<tr>
<th>pH Choice</th>
<th>pH 4</th>
<th>pH 7</th>
<th>pH 9</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td># Observations</td>
<td>Total Observations</td>
<td># Observations</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
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</tr>
<tr>
<td>pH 4 vs. pH 7</td>
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</tr>
<tr>
<td>pH 9 vs. pH 7</td>
<td>X</td>
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</tbody>
</table>

Sample Recording of Data:

- pH 4 vs. pH 7: X
- pH 9 vs. pH 7: X
**EXPERIMENT 2B: PROCEDURE (continued)**

**B. Effect of Metals**

Name of First Metal Tested: ____________________________________________

<table>
<thead>
<tr>
<th>Concentration of Treated Side (parts per million)</th>
<th># Worms Moving Toward:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control Side (Water)</td>
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<td>Treated Side</td>
</tr>
<tr>
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<td># Observations</td>
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<td>0.01</td>
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</tbody>
</table>

LOEC = Lowest Observed Effect Concentration

LOEC value for __________ is between __________ and __________ parts per million (ppm)
**S#8 – EXPERIMENT 2 – PREFERENCE AVOIDANCE BEHAVIOR DATA SHEET**

Date: _______________       Hour: _______________

Observers: ____________________________________________________________

**B. Effect of Metals**

Name of Second Metal Tested: __________________________________________

<table>
<thead>
<tr>
<th>Concentration of Treated Side (parts per million)</th>
<th># Worms Moving Toward:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control Side (Water)</td>
</tr>
<tr>
<td></td>
<td># Observations</td>
</tr>
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<td>0.01</td>
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</tbody>
</table>

**LOEC = Lowest Observed Effect Concentration**

LOEC value for ____________ is between __________ and __________ parts per million (ppm)
S#9 – EXPERIMENT 3 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES

This experiment is similar to Experiment 2, except soil is used instead of paper towel.

PROCEDURE:

A. Effect of Sand: Into one raceway, add enough sand (sandbox sand, not the “sand” provided in the ant farm box) as shown in Figure R below to fully cover the plastic surface and to be at a depth equal to the thickness of the raceway. Place the plastic divider widthwise into the sand so that it divides the sand into two equal sections. Move the divider side to side to create a small gap (approx. 1-1.5 cm) between the two sand sections.

Figure R: Diagrammatic View for Experiment 3 Methods

Temporary barrier to: 1) prevent water and chemical solution from mixing, and 2) create space to place worm. Barrier removed prior to placing worm in raceway.
S#9 – EXPERIMENT 3 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES

1. **Effect of pH:** Add \( \leq 30\) mL of distilled water to the sand on one side of the divider. Sand should be moist to damp but not soaking. Add a similar amount of pH 9 to the sand on the other side of the divider. Place the worm in the gap and record which side it chooses. Repeat for 4 different worms. Remove the pH 9 sand, dispose of it in a “Treated Soil” container, and wipe the section clean. Add clean, dry sand and add a volume of pH 4 or vinegar (weak acid, i.e., is partially dissociated in an aqueous solution). Repeat procedure and record observations.

2. **Effect of Metals:** Remove all sand from previous experiment and replace with fresh, clean, dry sand after rinsing and drying the raceway. Make all your serial dilutions of each of the 4 metal solutions. Label each test tube with the metal concentration, e.g., 1000 ppm Cu, 100 ppm Cu, 10 ppm Cu, etc. Starting with the **LOWEST** concentration first, repeat methods outlined in part A.a above using clean, dry sand for each new concentration for a total of 4 different worms/concentration. Once the worm has reacted to the metal solution, you do not need to test higher concentrations. Metals should be chosen such that all four metals are tested by the class but any one group does 1-2 metals.

**B. Effect of Potting Soil:** Into one raceway, add enough potting soil as shown below to fully cover plastic surface and to be at a depth equal to the thickness of the raceway. Place the plastic divider widthwise into the potting soil so that it divides the potting soil into two equal sections. Repeat the methods above except you are now using potting soil instead of sand. You will likely need to use more water to make the potting soil moist-damp than you needed for sand.

**C. Disposal of Treated Soil:** Sand or potting soil that was treated with distilled water can be reused. Sand or potting soil that was treated with acid, base, or metal solutions cannot be reused. However, it can be placed in a compost pile (a good place to also place your worms after the experiments are completed!). When diluted with other soil in the bin, the metals provide good nutrients for future plants.
S#9 – EXPERIMENT 3 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES DATA SHEET

Date: _______________       Hour: ____________________

Chemical: ________________________________   Soil Type: Sand

Observers: ______________________________________________________________

A. Effect of pH

   Acid Used: ____________________________________________

For each row, place a check in the box labeled # Observations each time you observe a worm choosing a specific pH value among the choices given in that row. Enter the number of total check marks for that pH choice in the box labeled Total Observations. An example is provided in the row labeled Sample Recording of Data.

<table>
<thead>
<tr>
<th>pH Choice</th>
<th>pH 4</th>
<th>pH 7</th>
<th>pH 9</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td># Obs</td>
<td>Total</td>
<td># Obs</td>
</tr>
<tr>
<td>Sample Recording</td>
<td>/</td>
<td>1</td>
<td>///</td>
</tr>
<tr>
<td>of Data</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>pH 4 vs. pH 7</td>
<td>X</td>
<td></td>
<td></td>
</tr>
<tr>
<td>pH 9 vs. pH 7</td>
<td>X</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
# S9 – EXPERIMENT 3 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES

Date: _______________       Hour: ________________

Chemical: ________________________________   Soil Type: **Sand**

Observers: ___________________________________________________________

**B. Effect of Metals**

<table>
<thead>
<tr>
<th>Concentration (parts per million)</th>
<th>Control Side (Water)</th>
<th>Treated Side</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td># Observations</td>
<td>Total</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
<td>/</td>
<td>1</td>
</tr>
<tr>
<td>1000</td>
<td></td>
<td></td>
</tr>
<tr>
<td>100</td>
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<tr>
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<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td></td>
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</tr>
</tbody>
</table>

Name of Second Metal Tested: _______________________________________________________

Chemical: ________________________________   Soil Type: **Sand**

<table>
<thead>
<tr>
<th>Concentration (parts per million)</th>
<th>Control Side (Water)</th>
<th>Treated Side</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td># Observations</td>
<td>Total</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
<td>/</td>
<td>1</td>
</tr>
<tr>
<td>1000</td>
<td></td>
<td></td>
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<td>1</td>
<td></td>
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<tr>
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<td></td>
<td></td>
</tr>
</tbody>
</table>
S#9 – EXPERIMENT 3 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES

Date: _______________       Hour: ____________________
Chemical: ________________________________   Soil Type: Potting Soil
Observers: ________________________________________________________________

A. Effect of pH

Acid Used: __________________________________

For each row, place a check in the box labeled # Observations each time you observe a worm choosing a specific pH value among the choices given in that row. Enter the number of total check marks for that pH choice in the box labeled Total Observations. An example is provided in the row labeled Sample Recording of Data.

<table>
<thead>
<tr>
<th>pH Choice</th>
<th># Worms Found In:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>pH 4</td>
</tr>
<tr>
<td></td>
<td># Observations</td>
</tr>
<tr>
<td>Sample</td>
<td></td>
</tr>
<tr>
<td>Recording</td>
<td></td>
</tr>
<tr>
<td>Data</td>
<td></td>
</tr>
<tr>
<td>pH 4 vs. pH 7</td>
<td></td>
</tr>
<tr>
<td>pH 9 vs. pH 7</td>
<td></td>
</tr>
</tbody>
</table>

Sample Recording of Data:

- pH 4 vs. pH 7:
  - X
- pH 9 vs. pH 7:
  - X
### S#9 – EXPERIMENT 3 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES

Date: _______________       Hour: _______________

Chemical: ________________________________   Soil Type: **Potting Soil**

Observers: ____________________________________________________________

#### B. Effect of Metals

Name of First Metal Tested: ____________________________________________

<table>
<thead>
<tr>
<th>Concentration (parts per million)</th>
<th># Worms Found In:</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control Side (Water)</td>
<td>Treated Side</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td># Observations</td>
<td>Total</td>
<td># Observations</td>
<td>Total</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
<td>/</td>
<td>1</td>
<td>///</td>
<td>3</td>
</tr>
<tr>
<td>1000</td>
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<tr>
<td>100</td>
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<tr>
<td>0</td>
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<td></td>
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<td></td>
</tr>
</tbody>
</table>

Name of Second Metal Tested: ____________________________________________

Chemical: ________________________________   Soil Type: **Potting Soil**

<table>
<thead>
<tr>
<th>Concentration (parts per million)</th>
<th># Worms Found In:</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control Side (Water)</td>
<td>Treated Side</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td># Observations</td>
<td>Total</td>
<td># Observations</td>
<td>Total</td>
</tr>
<tr>
<td>Sample Recording of Data</td>
<td>/</td>
<td>1</td>
<td>///</td>
<td>3</td>
</tr>
<tr>
<td>1000</td>
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<td>100</td>
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<td>10</td>
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<tr>
<td>1</td>
<td></td>
<td></td>
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<tr>
<td>0</td>
<td></td>
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</tr>
</tbody>
</table>
This experiment is similar to Experiment 3, except that the worm is burrowing first through a control soil before it encounters a treated soil. **BE SURE TO WEAR LAB GLOVES DURING THIS EXPERIMENT AS YOU WILL BE HANDLING CHEMICALS.**

**DISPOSAL:**
All used potting soil should be discarded into a specially designated bucket after all worms have been tested for each pH and metal concentration. Sand or potting soil that was treated with distilled water can be reused. Sand or potting soil that was treated with acid, base, or metal solutions cannot be reused. However, it can be placed in a compost pile (a good place to also place your worms after the experiments are completed!). When diluted with other soil in the bin, the metals provide good nutrients for future plants. Rinse and dry the ant farm between each experiment.

**PROCEDURE:** Refer to **Figure S** for a diagrammatic view for Experiment 4 methods.

**D. Effect of pH:**

1. Prepare a pH 4 solution using 15 mL of aluminum sulfate (Al$_2$(SO$_4$)$_3$) dissolved into 1 liter of distilled water. Use NaOH or KOH solution using distilled water to prepare pH 9. **Why not use dechlorinated water?**

2. Tape the divider that separates the two raceways so that the hole in the divider is blocked (Fig. S1). Tape the two pieces together so that the hole created when the two pieces are assembled is covered (Fig. S2a. Turn the assembly over and place a piece of masking tape 6 cm from the bottom (Fig. S2b). This will indicate to what level the bottom layer of will extend. Another piece of tape will be placed 10 cm above this mark and will indicate the volume for the top layer of soil. Mark the left side TREATED and the right side UNTREATED.

3. Into left raceway (TREATED), add approx. 15 ml of potting soil to the bottom half of the worm raceway (Fig. S3a). Keep it loosely packed so that it comes to a height of 6 cm from the bottom end of the raceway. If the soil is too tight, the worm will have difficulty burrowing and you will have difficulty seeing the worm in the raceway. Gently add 10 mL of either pH 4 or 9 to the soil. Be sure that it is evenly moist. Do the same procedure on the right half of the raceway (UNTREATED) except that you will add only distilled water. **WHY?**

4. Add 10 cm of potting soil to the top half of the raceway so that you now have 16 cm of soil (Fig. S3b). To this second layer of soil, add 5 mL of distilled water. Repeat on the other side of the worm raceway. **DO NOT LET THE TOP AND BOTTOM SOILS TOUCH EACH OTHER UNTIL BOTH HALVES HAVE BEEN TREATED WITH THEIR RESPECTIVE SOLUTIONS!! WHY?**

5. Using the pH soil test kit, measure the potting soil pH at 3 different locations on each side of the raceway before adding the worms and closing up the apparatus: bottom, top, and 1 cm above the location where the two soils meet. Record this information on Data Sheet A. **Why is this important?**

6. Place 2 worms onto the top layer of each raceway (2 on treated side and 2 on untreated side) and close up the ant farm. Clamp entire apparatus to a ring stand.
S#10 – EXPERIMENT 4 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES

7. Observe the worm movement over a 10 minute period. Record on Data Sheet B how far down (cm) the worm moved for each raceway.
8. Open the ant farm, remove the worms, and repeat procedure with 4 new worms. Continue until you have tested at least 6 worms for each pH.

E. Effect of Metals:
1. Procedure is the same as above except you will be using metal solutions.
2. Into left raceway (TREATED), add approx. 15 ml of potting soil to the bottom half of the worm raceway (Fig. S3a). Keep it loosely packed so that it comes to a height of 6 cm from the bottom end of the raceway. If the soil is too tight, the worm will have difficulty burrowing and you will have difficulty seeing the worm in the raceway. Gently add 5 mL of 1000 ppm metal solution to the soil. Be sure that it is evenly moist. Do the same procedure on the right half of the raceway (UNTREATED) except that you will add only distilled water. WHY?
3. Add 10 cm of potting soil to the top half of the raceway so that you now have 16 cm of potting soil. To this second layer of potting soil, add 5 mL of distilled water. Repeat on the other side of the worm raceway (Fig. S3b). DO NOT LET THE TOP AND BOTTOM SOILS TOUCH EACH OTHER UNTIL BOTH HALVES HAVE BEEN TREATED WITH THEIR RESPECTIVE SOLUTIONS!! WHY?
4. Place 2 worms onto the top layer of each raceway (2 on treated side and 2 on untreated side) and close up the ant farm. Clamp entire apparatus to a ring stand (Fig. S4).
5. Observe the worm movement over a 10 minute period. Record on Data Sheet B how far down the worm moved for each raceway.
6. Open the ant farm, remove the worms, and repeat procedure with 4 new worms. Continue until you have tested at least 6 worms for each treatment.
7. Repeat procedures (B2-B6) above except use a 100 ppm metal solution for the bottom layer on the treated side.
8. Again, repeat procedures (B2-B6) above except use 10 ppm and 1 ppm metal solutions.

F. Effect of soil type: Repeat entire protocol except use sand instead of potting soil.

NOTES:
3) To save class time, each pair of students can be assigned a specific comparison, e.g., 1000 and 100 ppm metal solution with potting soil. Have all students begin their experiment at the same time and then at the end of the 10-minute period, have students record their data on a common data form so all students can record the data from the entire class on their Data Sheet. In this way, over a 2-day period, the effect of pH, metals, and soil type can be observed and recorded by all the students.
4) If the worm has gone below 6 cm, it has entered the layer with the chemical contaminant. There may be some diffusion of the chemical into the top layer closest to the contaminated soil. If this happens, how would you determine this simply by observing the worm’s pattern of burrowing?
Optional Experiments: What other chemicals could contaminate your soil? How sensitive are worms to them?

Figure S:

5. Preparation of Worm Raceway

6. Marking Soil Levels on Worm Raceway
   a.                             b.

7. Adding Soil and Chemical Solutions
   a.                             b.

8. Assembled Apparatus

5. pH Soil Test Kit
S#10 – Experiment 4 – Effect of Soil Type on Behavior Responses - Data Sheet A

Date: ________________________   Hour: _________________________
pH: ________________________   Soil Type: ______________________
Observers: ____________________________________________________

<table>
<thead>
<tr>
<th>TREATED</th>
<th>UNTREATED</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH ____</td>
<td>pH ______</td>
</tr>
<tr>
<td>pH ____</td>
<td>pH ______</td>
</tr>
<tr>
<td>pH ____</td>
<td>pH ______</td>
</tr>
<tr>
<td>pH ____</td>
<td>pH ______</td>
</tr>
</tbody>
</table>
**S#10 – EXPERIMENT 4 – EFFECT OF SOIL TYPE ON BEHAVIOR RESPONSES-DATA SHEET B**

Date: ________________________   Hour: _________________________

Chemical: _________________________   Soil Type: _________________________

Observers: ______________________________________________________________

Name of metal tested: _____________________________________________________

<table>
<thead>
<tr>
<th>Worm #</th>
<th>Distance (cm) burrowed from top with bottom layer containing:</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>pH 4</td>
</tr>
<tr>
<td>1</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td></td>
</tr>
<tr>
<td>4</td>
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<tr>
<td>5</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td></td>
</tr>
<tr>
<td>Average</td>
<td></td>
</tr>
</tbody>
</table>

Observations:
Answer the following questions related to this experiment:

1. For each of the trials, why was it important to have one side of the raceway treated with only distilled water (labelled as “untreated”)?

___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________

2. For the pH experiment, why was it important to measure the soil pH at 3 different locations on each side of the raceway before adding the worms and closing up the apparatus?

___________________________________________________________________________
___________________________________________________________________________
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___________________________________________________________________________
___________________________________________________________________________

3. Was there a difference in burrowing depth for pH 4, pH 9, and the control side? If so, suggest a hypothesis for that difference.

___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________

4. Was there a difference in burrowing depth for 1000 ppm, 100 ppm, 10 ppm and 1 ppm of the metal solutions? If so, suggest a hypothesis for that difference.

___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________

5. Was there a difference in burrowing depth in sand vs. potting soil? If so, suggest a hypothesis for that difference.

___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
**S#11 – OBSERVING CATION EXCHANGE CAPACITY OF SOILS**

**PROCEDURE:**

A. By each balance, place 2 – 100 mL beakers, one of which will be filled 1/3 with potting soil and the other 1/3 with sand. Moisten the two soil types with distilled water but do not soak. Drain excess water.

B. On the acetate transparency, draw two 1 cm diameter circles with each circle 1 cm from the next circle. Place the petri dish over the two circles and place the petri dish and the transparency onto the dissecting scope stage. Adjust the focus so that you can see both circles simultaneously.

C. Weigh out 0.05g – 0.075 g of each soil type. Place each soil type in the Petri dish over one of the circles. Spread out so that the soil particles are as separated as possible but still within the 1 cm diameter circle. Repeat for the other soil type.

D. Add 1 drop of methylene blue to the potting soil and immediately start one of the stop watches. Then add 1 drop of methylene blue to the sand and immediately start the other stop watch. On the data sheet record the time (in seconds) for the blue color to visibly begin disappearing, and then the time at which all the blue color is gone for each soil type. Observe for a maximum of 5 minutes.

E. Repeat with any concentrated metal solution (1000 parts per million) that has a strong color, e.g., nickel or copper.

**Methylene blue**

<table>
<thead>
<tr>
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<th>Time (sec)</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Begin to Disappear</td>
</tr>
<tr>
<td><strong>Potting Soil</strong></td>
<td></td>
</tr>
<tr>
<td><strong>Sand</strong></td>
<td></td>
</tr>
</tbody>
</table>
### S#11 – OBSERVING CATION EXCHANGE CAPACITY OF SOILS

Chemical 1: ________________

<table>
<thead>
<tr>
<th></th>
<th>Time (sec)</th>
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</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Begin to Disappear</td>
<td>Disappear Totally</td>
<td></td>
</tr>
<tr>
<td>Potting Soil</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sand</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Chemical 2: ________________

<table>
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<th></th>
<th>Time (sec)</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Begin to Disappear</td>
<td>Disappear Totally</td>
<td></td>
</tr>
<tr>
<td>Potting Soil</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sand</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
S#11 – OBSERVING CATION EXCHANGE CAPACITY OF SOILS

Answer the following questions related to this experiment:

1. Which soil type absorbed the blue color faster? ________________________________

2. Why did you have to moisten the soil first?
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________

3. Develop a hypothesis to explain your results.
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________

4. Relate your hypothesis to the concept of cation exchange capacity.
___________________________________________________________________________
___________________________________________________________________________
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5. How do your results help to explain the changes in response by the earthworm to a chemical alone (Experiment 1) vs. in a soil (Experiment 3)?
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___________________________________________________________________________
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6. Compare and contrast the observations for the methylene blue and the metal solution.
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7. If you tried more than one metal solution, is there a difference in the rate of disappearance between them? Suggest a reason for the difference.
___________________________________________________________________________
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___________________________________________________________________________
Answer the following questions related to the experiments:

1. What does LOEC mean? In Experiment 1, at what pH did the worm react adversely? Develop an experiment that would allow you to calculate the LOEC for pH.

___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
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2. How does the LOEC differ between just adding drops of chemical on the plastic raceway vs. adding the chemical into either potting soil or sand? Provide a hypothesis that could explain these differences.

___________________________________________________________________________
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3. Compare the LOEC for each metal in potting soil vs. sand. If there is a difference, suggest an explanation. How might this idea be tested experimentally?

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4. Explain the concept of cation exchange capacity of soil. How does this change your hypothesis from question 3? Develop an experiment to test this new hypothesis.

___________________________________________________________________________
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5. Describe the worm’s “decision-making process” when placed between the water and chemically treated sides.

___________________________________________________________________________
___________________________________________________________________________
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___________________________________________________________________________
6. Describe the worm’s reaction (toward or away) from each chemical in each of the three experiments.
___________________________________________________________________________
___________________________________________________________________________
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___________________________________________________________________________

7. In Experiment 1, did the worm react similarly whether the worm moved forward or backward into the chemical? Develop a hypothesis to explain those observations that accounts for both the structure of the nervous system and ecological implications.
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________

8. Why does the data sheet want you to identify the LOEC as “between” rather than an exact value? What would you have to do to find the exact LOEC?
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________

9. What are the two types of acid rain? Develop an experiment that would test the hypothesis that the LOEC for both acid types is the same. Compare the LOEC of strong (e.g., HCl, H_2SO_4 or HNO_3) vs. weak acids (e.g., acetic acid [vinegar]).
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________
___________________________________________________________________________

10. If a concentration is below the LOEC level, does that mean it is not toxic to the worm? Defend your answer by providing logical reasoning and scientific evidence to support your opinion. Design an experiment to test your hypothesis.
___________________________________________________________________________
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INTRODUCTION

Environmental agents include substances in our surroundings that may affect living organisms. We are generally unaware of many of them because they are present in our foods, drinking water or air in very small amounts.

Environmental agents may occur naturally in the environment or be made by humans and introduced into the environment. Living things exposed to these agents may change noticeably, change ever-so-slightly or not change at all. Scientists now understand that the kind and severity of change in an exposed organism is often determined by how old the organism was at the time of exposure, how long the organism was exposed, and by how much of the substance the organism was exposed to. Commonly, the earlier an organism is exposed, the greater the effect of the agent.

NATURALLY OCCURRING ENVIRONMENTAL AGENTS

Naturally occurring agents include table salt, nicotine and alcohol. Many natural substances are required for normal development. Among the examples above, salt is the only essential substance. But when present in the body in amounts too large or small, even an essential agent like salt may cause health problems for humans. For example, too much salt may result in high blood pressure.

Harmful health effects are caused not only by inappropriate amounts of essential agents, but also by nonessential ones like nicotine and alcohol. Nicotine in cigarette smoke and chewing tobacco causes addiction. In turn, addiction to smoking leads to heart disease, emphysema, cancer and stroke. Alcohol presents its own set of health problems. For instance, when exposed to alcohol, a baby in the womb is at risk of developing life-long abnormalities of its nervous and skeletal systems. As a consequence of these harmful effects, women who are pregnant or wish to become pregnant are advised not to smoke cigarettes or drink alcohol.

SYNTHETIC ENVIRONMENTAL AGENTS

Unlike natural environmental agents, “synthetic” environmental agents are substances that have been made or are being made in chemical laboratories and factories. Tens of thousands of these synthetic agents have been introduced into our surroundings through their production, use and disposal. Like natural agents, synthetic agents may or may not be harmful.

Our exposure to most synthetic agents is involuntary; we do not choose to be exposed to them, but are exposed because their production, use and disposal are not isolated from the general environment. Manufacturers and consumers - homeowners, farmers, governmental agencies and business owners - are responsible for their presence in our surroundings. Through their production and sale, manufacturers introduce these agents into our environment. Consumers release them when they use and dispose of them. Because consumers use and dispose of
numerous synthetic substances – pesticides to kill insects, weeds or fungi; cleaning agents to remove dirt or kill bacteria or molds; and solvents to make, run or repair appliances, office equipment and production machinery - they are spread throughout our surroundings.

CONCLUSION

As a technical society, we have become dependent on synthetic environmental agents. However, many of these agents threaten our health and the health of our environment. The reading in this module, “A Is for Aldicarb, P Is for Pesticide,” focuses on one synthetic environmental agent of concern.
A. How does aldicarb move from the soil into an insect? A human?

B. What does aldicarb do to the nervous system of an insect? A human?

C. What evidence is there that some of the aldicarb that is absorbed into the plant may eventually be eaten by humans?

D. What happens to the aldicarb that is placed in the soil but does not enter the plant?

E. How does aldicarb get into the water we drink?

F. Using the information from the answers to the questions above, explain how aldicarb might get into our bodies and how it may affect our health.
Note to the Teacher: Take a few minutes and have student groups read and discuss the Introduction and look at the Concept Map. As a class, briefly discuss student responses to the questions and Concept Map. Then assign each group one of the six sections of the reading, instructing them to complete the appropriate section of the concept map and prepare to report to the class what they learned (The more difficult sections of the reading may be assigned to two groups so checks on understanding come mostly from classmates.) Once groups are finished, have each group report to the class summarizing its section of the reading. Close the class discussion with a review of the completed Concept Map.

INTRODUCTION

In the reading that follows, you will learn about a chemical pesticide called aldicarb. The reading begins with basic information about pesticides. It then discusses how aldicarb is used, how it works, its regulation, and why people are concerned about aldicarb — as well as other pesticides used on food crops. The conclusion discusses how aldicarb has been regulated with the goal of preventing harm to humans.

Before you begin reading about aldicarb, think about and answer these questions:

1. How do insects react when you spray them with a pesticide like Raid™ or Spectracide™?
2. If sprays like Raid™ or other insecticides can kill insects in a matter of a few minutes, how strong or powerful are the chemicals used in the spray?
3. Could the health of a bird or other organism be affected if it were to eat many insects treated with a pesticide?

On the next page you will see a concept map covering the content of the reading on aldicarb. Some of the information has been filled in for you. As you read the article add information to the map in the spaces provided.
1. Pesticides may be thought of as environmental agents. Environmental agents include physical agents like heat and light, chemical agents like salt and pesticides, and biological agents like bacteria, worms, elephants and molds. Our surroundings are full of environmental agents that might affect us and other living organisms — for good or ill.

2. Pesticides are chemical or biological agents used to kill pests like insects, mice, rats, and molds. Regardless of its target pest, the purpose of a pesticide is to kill the pest without harming the species being treated or “protected.” However, a pesticide may affect organisms other than those being treated or killed. Whether or not it affects other organisms depends on the make-up of the pesticide (whether it is designed to kill one kind of pest or many kinds of pests), how long the pesticide stays in the environment, and how it is transferred to the pest. Pesticides are delivered to pests in three main ways: sprayed directly on the pest (like many insecticides), presented in a food eaten by the pest (like ant traps or rat bait), or taken up by a living organism as it feeds on another living organism that has taken the pesticide into its body. Aldicarb is a pesticide of the last type. The pests eat plants that have taken in aldicarb as their roots take up water. Pesticides are among the most regulated environmental agents in the United States. Several federal governmental agencies regulate pesticides — their manufacture, distribution, use and environmental presence. The Federal agencies that regulate pesticides include the Environmental Protection Agency (EPA), the Food and Drug Administration (FDA), and the U.S. Department of Agriculture (USDA). These agencies are authorized by the U.S. Congress to enforce more than 14 separate regulations that govern pesticides. They are responsible for protecting public health and regulating materials that present possible hazards to humans or the environment. Aldicarb is one of many highly effective pesticides. Because it presents hazards to humans, it is regulated by several agencies.

3. Aldicarb Characteristics and Its Use on Plants
   Aldicarb is a chemical used around the world to kill plant pests that pierce and suck on plant leaves, stems, and roots. It contains five elements (carbon, hydrogen, oxygen, nitrogen and sulfur), is a white solid, and is extremely poisonous. It dissolves readily in water, but is not sprayed on crops because it could easily harm a person applying the pesticide. Instead, farm workers drill the highly toxic pesticide into the ground as a grainy powder. Because it is so toxic, by law only a trained, certified person may apply the pesticide. And only after a number of days have passed may a person enter a treated field.

   As the certified farm worker plants a row of seeds or treats a growing crop, a granular powder containing aldicarb is drilled about 5 to 7 centimeters into the soil near the seeds or plants. With the next rainfall, the aldicarb dissolves and plant roots take up the aldicarb-laced water. Once in the plant, the plant changes aldicarb into “metabolite” pesticides that are even more toxic than the aldicarb. As the plant sap moves throughout the plant, the aldicarb and its metabolites are distributed to all plant parts — roots, stems, leaves, blossoms and fruits. When small sucking organisms like mites, nematodes, and insects (aphids, whiteflies, leaf miners and others) pierce the plant tissue, they take in the plant sap containing the dissolved pesticides and are killed.
Because aldicarb and its metabolites stay in the plant for some time, feeding pests continue to die. As long as aldicarb and its metabolites are in the plant, the crop is protected from pests, and people who pick and eat the crop are at risk.

4. Government Regulation of Aldicarb Based on Scientific Studies

In an attempt to protect people from the harmful effects of aldicarb and its metabolites, governments regulate not only how aldicarb is applied to a crop, but also how long a grower must wait to harvest an aldicarb-treated food crop. In the United States, different food crops have different harvest wait times. For example, a grower must wait 30 days after treatment to harvest lemons, 120 days to harvest sugarcane, and 120 days to harvest sweet potatoes.

To set regulations like these, governmental officials rely on scientific studies. The studies include the best time to apply aldicarb to a particular crop, how much aldicarb can be used for that crop, how long aldicarb stays in different kinds of soil after being applied, and how long it takes a particular kind of plant to change aldicarb into harmless substances. As time passes, the amount of aldicarb (and its metabolites) in the plant will decrease. But some pesticide may remain at harvest time. Rules about application amounts, times and procedures are used in an attempt to protect people from eating food crops that might contain harmful amounts of aldicarb.

Based on scientific data, governments around the world make rules about the use of aldicarb. However, not all countries set the same rules and health protection standards because conditions are different around the world. And not all governments have the will and/or ability to check that all growers obey the rules. This means some crops treated with aldicarb may contain more aldicarb (and metabolites) than is allowed by government regulations.

Food crops are not the only way people are exposed to aldicarb. It may also be in their drinking water. Runoff from rainfall carries some of the pesticide into rivers, streams and lakes. These surface waters are the source of drinking water for many people in the United States. In some areas of the nation, rainfall that soaks into the ground carries pesticide into groundwater that supplies the wells of many small towns and individual farms. Because aldicarb has contaminated some drinking water supplies, our national government has set a standard for how much aldicarb may be in our drinking water. The standard is now set at 0.003 mg of aldicarb/L (or 0.000003g/L) of drinking water. The goal for a future drinking water standard is 0.001 mg/L (or 0.000001g/L). But because the foods we eat may also be a source of aldicarb, the government has ruled that the total amount of aldicarb an adult may eat and drink in one day is 0.004 mg/kg body weight (or 0.000004g/kg). This standard means a 120 pound adult may eat and drink up to 0.220 mg (or 0.00022 grams) of aldicarb in one day.
5. Uses of Aldicarb and How It Works
Since 1970, aldicarb has been used to protect a variety of crops and help growers have plentiful harvests. Aldicarb is used mainly on cotton; other treated crops include bananas, grapes, grapefruit, hops, lemons, lettuce, oranges, peanuts, pecans, sorghum, soy beans, strawberries, sugar beets, sugarcane, sweet potatoes, tomatoes and decorative plants. Because too much of the pesticide was found in potatoes grown in the United States, its use on potato crops ended in the United States in 1987. However, because it works very well, aldicarb is still used to protect many crops from pests.

Aldicarb works well because it kills pests by upsetting the way their nerves signal muscles. Normally, nerves send an electrical message or impulse down the length of a nerve cell. When the impulse reaches the end of the nerve cell, a chemical is released at the end of the nerve cell. The chemical relays the nerve message across a small space to a muscle cell. In this way the muscle cell gets a signal to do its job, namely contract. To keep the muscle from being over-worked, a key enzyme destroys the chemical messenger. With the chemical messenger destroyed, the muscle cell relaxes and is able to receive the next message and do its job again. Aldicarb upsets this system. (See “Nerve-Muscle Interactions” C#2.)

Specifically, aldicarb keeps the enzyme from destroying the chemical messenger so the chemical messenger keeps on stimulating the muscle cell. This causes the muscle to repeatedly contract until it gets so tired it will not work. To feel what it is like to contract a muscle, make a tight fist — hold it for a few minutes. How do the muscles in your hand feel? If the particular muscles involved are used for breathing, those muscles will stay contracted and breathing will stop. The organism will die from a lack of oxygen. Aldicarb is a most effective pesticide because it affects other insect nerve pathways in the same way it affects the breathing pathway. All nerve pathways of a sucking insect looking for food are disrupted. The insect dies and is unable to reproduce. Aldicarb does an excellent job of killing pests, therefore disrupting nerve-muscle signaling in a host of species.

6. Aldicarb Use on Food Crops: A Reason for Concern
Aldicarb is of concern to people because in small amounts it acts on our nervous systems in the same way as it acts on those of insects. Aldicarb upsets our nerve messages by keeping the key enzyme from breaking down the chemical messenger. A person exposed to a very high amount of aldicarb may die or may arrive at an emergency room with a very slow heart rate or in a coma. With proper treatment, emergency room patients usually survive. People who have been exposed to aldicarb at a level higher than permitted (total daily intake of 0.004mg/kg of body weight) may experience dizziness, nausea, diarrhea, sweating, blurred vision, tiny pupils, difficulty breathing, shaky muscles and leg weakness. Usually these exposed individuals pass most of the aldicarb out of their body within 24 hours. However, the effects of long term exposure to low levels of aldicarb are unclear. Aldicarb has not been tested enough to state whether or not it or its “metabolites” might cause diseases of the nervous system or cancer due to eating contaminated food or drinking contaminated water.
In scientific experiments, earthworms exposed to amounts present in treated soil grow more slowly and have less body protein. Frogs exposed to a pesticide mixture including aldicarb showed a lowered immune response to a frog parasite. Women regularly exposed to aldicarb showed abnormal populations of a type of white blood cell (T-cell) that protects from body invaders. Very low doses of aldicarb given to pregnant rats affected the fetuses more than the mother; and pregnant rats kept the aldicarb in their bodies longer than did rats that were not pregnant. Yet three generations of rats exposed to higher doses showed no negative effects. Finally, in human cell cultures, aldicarb caused mutations and various kinds of damage to chromosomes.

7. Aldicarb in Wisconsin
In the 1970s and early 1980s, aldicarb was regularly used on potato crops in Wisconsin. In 1978, toxic wastes came to the surface of the ground in residential neighborhoods at Love Canal, New York. The event prompted people around the U.S. to check groundwater for contaminants. Studies soon documented many wells were contaminated with agricultural chemicals or materials that had moved out of landfills and dumps into the groundwater. In Wisconsin, Dr. Byron Shaw of the University of Wisconsin – Stevens Point discovered aldicarb in Wisconsin wells. His report prompted further testing of wells in areas where aldicarb was used. When aldicarb was found in more wells, the State Legislature formed a committee led by State Rep. Mary Lou Munts and a bill was drafted. By May, 1984, Wisconsin Governor Tony Earl signed the state’s groundwater protection law. It assured that all groundwater in Wisconsin would be protected. Rules for using aldicarb (ATC 30.24) were established in Wisconsin; these rules limit how much aldicarb can be used and require growers to inform the Wisconsin Department of Agriculture, Trade and Consumer Protection before they use it. Today, no one in Wisconsin is using aldicarb even though it could be used on crops like soybeans. Anyone using aldicarb in Wisconsin must obey the state rules that limit its use to 2 pounds of aldicarb per acre per year – every other year on a particular field. Wisconsin’s strict rules are meant to protect our food and drinking water from contamination, but not all our food and water comes from Wisconsin. We must therefore rely on national and international rules to protect us from harm caused by specific environmental agents.
**Note to the Teacher:** This exercise will help student pairs or groups analyze both normal and aldicarb-affected nerve-to-muscle signaling. Orient students to the illustrations below.

1. **Using the key below, have students enter the name of each structure described in the first column (orange) in the table below.**
2. **Have students discuss and record the changes in appearance and position of each structure as one moves from a to b to c in the middle row of C#2.**
3. **Follow with a class discussion that clarifies what the changes mean for normal nerve-to-muscle signaling. Using this approach, have students analyze the effect of aldicarb on nerve-to-muscle signaling.**

![Diagram of aldicarb-affected nerve to muscle signaling at the synapse](source: University of Wisconsin-Milwaukee)

This diagram illustrates the sequence of events at a nerve-muscle synapse affected by the environmental agent aldicarb.

a) Chemical messenger (green half-capsules) is released at the end of the nerve axon (light brown) into the synapse (gap) between the nerve ending and the receiving, relaxed muscle (pink showing less overlap between filaments) with receptor sites (yellow).

b) Chemical messenger reacts with the receptor sites and stimulates the muscle to contract (overlap between filaments increases and muscle contracts).

c) The enzyme associated with the synapse (light blue arcs on the muscle) is inhibited by the environmental agent (dark blue arcs over light blue arcs) preventing the break-down of the chemical messenger and the muscle stays contracted (over lap between filaments stays increased). Contracted muscles cannot do the job for which they were intended; namely, contract to move a body part – engaged in lifting, walking, moving the eyes in their sockets, moving food through the gut, or breathing. Aldicarb’s effectiveness is a result of its ability to upset normal nerve-muscle signaling at the synapse, resulting in the inability of muscles to do this work. Aldicarb causes the death of insects that suck on plants as well as risk of illness or death in people who happen to ingest aldicarb.
**S#16 – HOW DOES ALDICARB WORK?**

**STUDENT DIRECTIONS:**

1. Using the key at the bottom of the illustrations about nerve-to-muscle signaling, enter the name of each structure listed in the first column below.
2. Look at the middle row of illustrations about normal signaling and beginning at “a” enter a description of each structure’s appearance and position.
3. Look at “b” and describe each structure’s appearance and position.
4. Finally, look, at “c” and describe the appearance and position of each structure.
5. With your group, review and discuss the changes from “a” to “b” and then “b” to “c.”
6. Repeat this process for aldicarb-affected nerve-to-muscle signaling as shown in the bottom row of illustrations of C#2.

### NORMAL NERVE-TO-MUSCLE SIGNALING

<table>
<thead>
<tr>
<th>STRUCTURES</th>
<th>(A)</th>
<th>(B)</th>
<th>(C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>GREEN CAPSULE</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Structure:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>YELLOW OVAL</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Structure:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PINK/GREY STRINGS</td>
<td></td>
<td></td>
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<tr>
<td>Structure:</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
## ALDICARB-AFFECTED NERVE-TO-MUSCLE SIGNALING

<table>
<thead>
<tr>
<th>STRUCTURES</th>
<th>(A)</th>
<th>(B)</th>
<th>(C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>GREEN CAPSULE</td>
<td></td>
<td></td>
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<tr>
<td>Structure:</td>
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<td>YELLOW OVAL</td>
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<td>Structure:</td>
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<tr>
<td>PINK/GREY STRINGS</td>
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<tr>
<td>Structure:</td>
<td></td>
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</tr>
</tbody>
</table>

What structure in the illustrations represents the aldicarb? Where is it found? What does aldicarb do?
Note to the Teacher: The video showing aldicarb’s effect on earthworm burrowing also illustrates nerve-to-muscle signaling. (Exercise S#12 relates to normal and aldicarb-affected signaling and is provided to facilitate student understanding of the affected behaviors observed in the video.).

PROBLEM:
Does Aldicarb Affect Earthworm Burrowing Time?

HYPOTHESIS:

EXPERIMENT:
This experiment has been done in an approved laboratory; laws will not allow aldicarb to be used in classrooms.

OBSERVATIONS/RESULTS:

**TABLE OF BURROWING TIMES FOR ALDICARB-TREATED EARTHWORMS**

As you watch the earthworm burrow, enter in the appropriate box the length of time (in minutes and seconds) it takes the worm to burrow out of sight.

<table>
<thead>
<tr>
<th>TOTAL TIME TO BURROW</th>
<th>CONTROL – 0 PPM</th>
<th>1.0 PPM</th>
<th>5.0 PPM</th>
</tr>
</thead>
<tbody>
<tr>
<td>EXPOSURE TIME</td>
<td>2.5 MIN</td>
<td>2.5 MIN</td>
<td>2.5 MIN</td>
</tr>
<tr>
<td></td>
<td>5 MIN</td>
<td>5 MIN</td>
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<tr>
<td></td>
<td>10 MIN</td>
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<td>20 MIN</td>
<td>20 MIN</td>
<td>20 MIN</td>
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<tr>
<td></td>
<td>10.0 PPM</td>
<td></td>
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</tr>
<tr>
<td>EXPOSURE TIME</td>
<td>2.5 MIN</td>
<td></td>
<td></td>
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<td></td>
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<tr>
<td></td>
<td>20 MIN</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>TOTAL TIME TO BURROW</th>
<th>20.0 PPM</th>
</tr>
</thead>
<tbody>
<tr>
<td>EXPOSURE TIME</td>
<td></td>
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<tr>
<td></td>
<td>2.5 MIN</td>
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<td></td>
<td>10 MIN</td>
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<td></td>
<td>20 MIN</td>
</tr>
</tbody>
</table>
S#17 – BURROWING BEHAVIOR OF ALDICARB-EXPOSED EARTHWORMS

OBSERVATIONS OF BURROWING OF ALDICARB-TREATED EARTHWORMS

As you watch the earthworms burrow, enter their burrowing times and describe their movements in the space below.
DIRECTIONS:

Select a color for the selected exposure time(s). Color the appropriate circle(s) in the Key below. Make a narrow bar graph for the selected concentration of aldicarb using the colors for the select exposure time(s). Then, using the same color, draw a line connecting the top of the 10 minute bars. Repeat the process for the other selected exposure time(s). Then answer the questions on the next page.

<table>
<thead>
<tr>
<th>COLOR KEY</th>
<th>EXPOSURE TIME</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0 Minutes</td>
</tr>
<tr>
<td></td>
<td>2.5 Minutes</td>
</tr>
<tr>
<td></td>
<td>5 Minutes</td>
</tr>
<tr>
<td></td>
<td>10 Minutes</td>
</tr>
<tr>
<td></td>
<td>20 Minutes</td>
</tr>
</tbody>
</table>

KEY: Color the circle for each exposure time
S#17 – BURROWING BEHAVIOR OF ALDICARB-EXPOSED EARTHWORMS

QUESTIONS: EFFECT OF ALDICARB ON EARTHWORM BURROWING TIME

1. Does exposure to aldicarb affect the time it takes an earthworm to burrow? Explain your answer by referring to the experimental results.

________________________________________________________________________
________________________________________________________________________
________________________________________________________________________
________________________________________________________________________
________________________________________________________________________

2. Is there any concentration and exposure time at which you saw no effect of aldicarb? Explain your answer by referring to the experimental results.

________________________________________________________________________
________________________________________________________________________
________________________________________________________________________
________________________________________________________________________
________________________________________________________________________

3. Do you think the results of this experiment justify scientists’ concerns about using aldicarb on food crops? Explain your answer based on the experiment and the reading about aldicarb (S#7).

________________________________________________________________________
________________________________________________________________________
________________________________________________________________________
________________________________________________________________________
________________________________________________________________________
AS YOU PLAN YOUR EXPERIMENT REMEMBER TO INCLUDE THESE PARTS OF AN INVESTIGATION:

- Problem Question
- Hypothesis
- Experiment
- Results/Observations
- Analysis of Results
- Conclusion

An analysis of the results often comes before the conclusion. The analysis can be the most interesting part of a report since it discusses different possibilities for explaining results (including scientific errors) and often leads to new questions and hypotheses. Below are details that will help you as you plan each part of your investigation. When your group believes it has done each item, check it and go on to the next item. (You probably will find the need to add to an earlier step as you work on a later step.) When you are done, if time permits, have another group check your plan. Finally, have your teacher check your plan and assign who will provide each piece of equipment.

**PROBLEM QUESTION:**

1. Clearly put into words the problem you are trying to solve. Do this in one sentence.
2. Test only one factor in your experiment and be sure the problem question deals with that factor.
3. Make sure you can work to solve the problem question over a period of two or three class days. Remember, well cared for worms will “work” while you are in other classes and at home!

**HYPOTHESIS:**

1. Write an answer to the problem question. It should be an educated guess based on your experiences and literature search.
2. Express your hypothesis in the future voice, for example, “Earthworms treated with _____ for __, __, and __ minutes will ______________.”
3. Remember, your hypothesis does not change based on your results; once the experiment is done you will evaluate your hypothesis – tell whether it is correct or incorrect – based on your results. A hypothesis that proves untrue is acceptable!
S#18 – GUIDELINES FOR PLANNING YOUR EXPERIMENT

EXPERIMENT:
((Some activities you have already done contain these sections. Use them as models for your work.)

1. Write a Materials section that lists all the equipment and worms you will need.
2. Decide which equipment group members will provide and what you think your school will provide. Agree as to who will provide each item.
3. Write a numbered, step-by-step procedure that any person could follow if they wanted to repeat your experiment and check your results.

RESULTS/OBSERVATIONS:

1. Imagine all the things you are likely to observe. List them and plan space for recording them. You may want to make tables, graphs and leave space for drawings.
2. If you expect to get quantitative data (numbers) be sure your table provides for that data. (Hint: Use the table provided for the video on aldicarb-exposed earthworms in S#16 as an example.)
3. If you expect to make qualitative observations, provide space for writing what you might see.
4. Record your results so they are easy to locate and use.

ANALYSIS:

1. Write what you thought you would observe, why you thought you would make that observation, and whether you indeed did observe the expected.
2. If you did not observe what you expected to, tell what you did observe and why you think you observed the unexpected.
3. List any experimental errors you may have made and explain how they might have affected the results and how you might avoid them.
4. List any new questions one might investigate after doing this experiment.
5. Propose an hypothesis for each question you have listed.

CONCLUSION:

1. Write a statement that answers the problem question.
2. Base that statement on the results/observations of your experiment.
3. Evaluate your hypothesis – tell whether it was correct or incorrect, or whether you have enough information to evaluate your hypothesis!
BACKGROUND

If humans were unable to learn they would not be able to walk, talk and hold jobs – skills needed for survival. To learn these skills, we depend on a large brain, but with brains the size of a small pin head, you might wonder if the earthworm’s survival depends on its ability to learn. You have studied the earthworm’s crawling behavior and “startle response.” You will now test to see if earthworms can learn by “habituation,” one kind of learning typical of many organisms. In these experiments, habituation means the earthworm will not exhibit the “startle response.”

Every day humans habituate to sensory stimuli; for example, traffic noises. After hearing the noises from a busy street for a while, a person no longer hears the sounds of passing traffic – the individual has learned to ignore the sounds. In this experiment with the earthworm you will watch for a change in its response to a repeated sensory stimulus (like a touch) – even if the stimulus prompts the “startle response.”

In its natural soil environment, the worm would rapidly burrow into the soil to avoid touch or capture by a bird or fisherman. The next time someone is digging in your garden, try to catch a worm while it is in its native environment to convince yourself that the worm is a good escape artist. In this experiment, the worm is on a flat surface into which it cannot burrow, so you will be able to repeatedly touch the worm and provide it with the opportunity to learn.

Test I – PROBLEM: CAN AN EARTHWORM LEARN?

HYPOTHESIS (WRITE WHAT YOU THINK WILL HAPPEN):

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________
EXPERIMENT:

MATERIALS (PER GROUP FOR TESTS I, II, AND III)

- Water
- Clean cloth or sponge
- Waxed paper cup
- 3 earthworms at room temperature
- Moist paper towel
- Your activity sheet and pencil
- Dropper bottle of dechlorinated tap water (to keep worms moist)
- A clean, well-rinsed surface – a table, lab bench, or area of the floor
- Stopwatch or watch that displays tenths of a second

PROCEDURE:

Before beginning clean and rinse your work area well. Have all materials available and choose roles: Timer, Experimenter and Recorder. All will later record.

REMEMBER: KEEP YOUR WORMS DAMP TO KEEP THEM FROM DRYING OUT AND SUFFOCATING.

1. **Experimenter**: Put one worm on the clean, well-rinsed surface.

2. **Timer**: Practice starting and stopping the stopwatch or reading seconds on a watch. When you are ready, tell the experimenter.

3. **Experimenter**: Stimulate the worm by touching its head. Do this by holding your hand about six inches away from the worm. Move your hand in to stimulate the worm with your thumb. Apply pressure like you would press the button of a remote control and then move your hand away. If the worm did not rapidly move away from your thumb, the timer should let you know when 15 seconds have gone by, then you should try again, using a slightly increased pressure. Once the animal gives a definite response, the timer should call out every 15 seconds and the experimenter should apply another stimulus.

4. **Recorder**: After the worm begins to respond, keep track of each stimulus on Table I. When the worm stops responding to three stimuli in a row, the animal has habituated to the stimulus – it has learned!

5. **All**: Enter the Recorder’s information in Table I, respond to questions 1 & 2, and write your “Conclusion” for this part of the activity.
S#19 – CAN EARTHWORMS LEARN?

Test I – PROBLEM: CAN AN EARTHWORM LEARN? (CONTINUED)

OBSERVATIONS/RESULTS I:

TABLE I

<table>
<thead>
<tr>
<th>STIMULUS</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>9</th>
<th>10</th>
<th>11</th>
<th>12</th>
</tr>
</thead>
<tbody>
<tr>
<td>“Startle Response” (Y/N)</td>
<td></td>
<td></td>
<td></td>
<td></td>
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<td></td>
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</tr>
</tbody>
</table>

1. Was the earthworm able to learn? ______ If so, what did it learn and what is the evidence that indicates it did learn?

   ______________________________________________________________
   ______________________________________________________________
   ______________________________________________________________

2. Is the ability to learn important for earthworms? ______ Explain:

   ______________________________________________________________
   ______________________________________________________________
   ______________________________________________________________

CONCLUSION I:

(Answer the problem question providing evidence from your activity and tell whether your hypothesis was correct or incorrect. Use complete sentences.)

Can an earthworm learn? ________ Explain:

   ______________________________________________________________
   ______________________________________________________________
   ______________________________________________________________
S#19 – CAN EARTHWORMS LEARN?

Test II – PROBLEM: CAN AN EARTHWORM RELEARN A SKILL - FASTER THAN IT FIRST LEARNED THAT SKILL?

HYPOTHESIS (WRITE WHAT YOU THINK WILL HAPPEN):

__________________________________________________________________________

__________________________________________________________________________

__________________________________________________________________________

EXPERIMENT:

MATERIALS (SEE TEST I MATERIALS)

PROCEDURE

1. Stop stimulating the worm for five minutes after it has habituated. Then apply the touch stimulus. (If the worm doesn’t respond, it has not forgotten what it learned; it is still habituated. Wait two more minutes and stimulate again. When the worm exhibits the “startle response,” it has forgotten what it learned and is no longer habituated. Can the worm relearn more quickly than it first learned?

2. Switch roles. (See Procedure, Test I.)

3. Repeat Steps 3 and 4 in Test I.

4. All: Enter the information in Table II, respond to items 1- 5, and write your “Conclusion” for this part of the activity.

OBSERVATIONS/RESULTS II:

TABLE II

<table>
<thead>
<tr>
<th>STIMULUS</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>9</th>
<th>10</th>
<th>11</th>
<th>12</th>
</tr>
</thead>
<tbody>
<tr>
<td>“Startle Response” (Y/N)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
</tbody>
</table>

1. Can an earthworm forget a learned behavior? _____ Provide evidence for your answer:

__________________________________________________________________________

__________________________________________________________________________

__________________________________________________________________________
2. Can an earthworm relearn a behavior? Provide evidence for your answer:

___________________________________________________________________________

___________________________________________________________________________

3. Check Tables I and II for the following information and record it below. Learning took _____ stimuli in Test I. Relearning took _______ stimuli in Test II. Compare and explain your answers.

___________________________________________________________________________

___________________________________________________________________________

4. Would habituation always be good for an earthworm? Explain your answer with an example.

___________________________________________________________________________

___________________________________________________________________________

___________________________________________________________________________

CONCLUSION II:

(Answer the problem question providing evidence from your activity and tell whether your hypothesis was correct or incorrect. Use complete sentences.)

1. Can an earthworm relearn a skill and relearn it faster than it first learned that skill? Explain:

___________________________________________________________________________

___________________________________________________________________________

___________________________________________________________________________

___________________________________________________________________________
S#19 – CAN EARTHWORMS LEARN?

TEST III – IS AN EARTHWORM HABITUATED TO A TOUCH ON ITS HEAD ALSO HABITUATED TO A TOUCH TO ITS TAIL?

HYPOTHESIS (WRITE WHAT YOU THINK WILL HAPPEN):

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

EXPERIMENT:

MATERIALS (SEE TEST 1)

PROCEDURE:

1. Switch roles. (See Procedure, Test I)

2. Repeat Steps 3 and 4 in Test I, but this time touch the worm’s tail, not its head.

3. **All**: Enter the Recorder’s information in Table III, respond to the question below and write your “Conclusion” for this part of the activity.

OBSERVATIONS/RESULTS III:

**TABLE III**

<table>
<thead>
<tr>
<th>STIMULUS</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>9</th>
<th>10</th>
<th>11</th>
<th>12</th>
</tr>
</thead>
<tbody>
<tr>
<td>“Startle Response” (Y/N)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1. How did the habituated earthworm respond to a touch to its tail? Do your results demonstrate that the earlier learning involved the worm’s entire body?

________________________________________________________________________

________________________________________________________________________

2. Would this be helpful to the earthworm? Explain:

________________________________________________________________________

________________________________________________________________________
CONCLUSION III:

(Answer the problem providing evidence from your activity and tell whether your hypothesis was correct or incorrect. Use complete sentences.)
Is an earthworm that is habituated to a touch on its head also habituated to a touch to its tail? Explain:

__________________________________________________________________________________
__________________________________________________________________________________
__________________________________________________________________________________
__________________________________________________________________________________

ANALYSIS:

1. What does this set of activities demonstrate about the earthworm’s ability to respond to stimuli in its environment?
__________________________________________________________________________________
__________________________________________________________________________________
__________________________________________________________________________________

2. How is its ability to sense stimuli in the environment important to the earthworm?
__________________________________________________________________________________
__________________________________________________________________________________
__________________________________________________________________________________
S#19 – CAN EARTHWORMS LEARN?

EXTENSIONS

A. Do you think the worm would remember as quickly if you waited a longer time (one hour or one day) before applying a stimulus the second, third, etc., time? If you want to try this experiment, put the habituated worm in a separate marked container with soil and set it aside for an hour or a day. Then repeat Test I above.

B. Test IV below.

TEST IV - IS AN EARTHWORM THAT HAS BEEN HABITUATED TO ONE KIND OF STIMULUS (TOUCH) ALSO HABITUATED TO A DIFFERENT KIND OF STIMULUS? (HEAT, CHEMICALS, LIGHT)

HYPOTHESIS (WRITE WHAT YOU THINK WILL HAPPEN):

________________________________________________________________________

________________________________________________________________________

EXPERIMENT:

MATERIALS:

___ Water
___ Clean cloth or sponge
___ Waxed paper cup
___ 3 earthworms at room temperature
___ Moist paper towel
___ Your activity sheet and pencil
___ Dropper bottle of dechlorinated tap water (to keep worms moist)
___ A clean, well-rinsed surface – a table, lab bench, or area of the floor
___ Dropper bottle of 7% ethyl alcohol (for test “a” below)
___ Alcohol lamp and glass rod (for test “b1” below)
___ Flashlight with intense narrow beam [maglight™] (for test “b2” below)
S#19 – CAN EARTHWORMS LEARN?

TEST IV - IS AN EARTHWORM HABITUATED TO ONE STIMULUS (TOUCH) ALSO HABITUATED TO A DIFFERENT KIND OF STIMULUS? (CONTINUED)

PROCEDURE

1. Habituate a worm to a touch on its head by repeating Test I.

2. Test the worm only one time using one of the following to answer the problem question:
   A. Chemical stimulus using ethyl alcohol: Using a dropper bottle of 7% ethyl alcohol, put a single drop of alcohol on the worm’s head. Did this stimulate a “startle response”?
   B. Physical stimuli
      1. Heat: Heat a glass rod in a cool flame so it gets hot but does not melt. Bring the rod close to the head of the worm but do not touch the worm’s skin. Does this stimulate a “startle response”?
      2. Light: Suddenly shine an intense beam of light on the worm’s head. Does this stimulate a “startle response”?

3. Enter data on Table IV. Then respond to items 1 thru 3 in the Analysis section.

4. Write your “Conclusion” for this part of the activity.

OBSERVATIONS/RESULTS IV:

TABLE IV

<table>
<thead>
<tr>
<th>STIMULUS</th>
<th>ALCOHOL</th>
<th>HEAT</th>
<th>LIGHT</th>
</tr>
</thead>
<tbody>
<tr>
<td>“Startle Response” (Y/N)</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

CONCLUSION IV:

Is an earthworm habituated to one stimulus (touch) also habituated to a different stimulus (chemical or physical?) Explain:
S#19 – CAN EARTHWORMS LEARN?

TEST IV - IS AN EARTHWORM HABITUATED TO ONE STIMULUS (TOUCH) HABITUATED TO A DIFFERENT KIND OF STIMULUS? (CONTINUED)

ANALYSIS:

1. If an earthworm learns that one stimulus (a touch on its head) is not harmful, has it learned other stimuli are not harmful? Explain using the results from Test IV.

________________________________________________________________________________________
________________________________________________________________________________________
________________________________________________________________________________________
________________________________________________________________________________________

2. What does the earthworm’s response to the chemical and physical stimuli tell you about its ability to detect different kinds of environmental stimuli?

________________________________________________________________________________________
________________________________________________________________________________________
________________________________________________________________________________________
________________________________________________________________________________________

3. How is sensitivity to different environmental stimuli important to the earthworm?

________________________________________________________________________________________
________________________________________________________________________________________
________________________________________________________________________________________
________________________________________________________________________________________
**Note to the Teacher:** You may wish to provide copies of pesticide labels. For safety reasons, do not bring products into the classroom.

You are shopping with your family at a garden store to buy what is needed for your vegetable garden. You think you might want to buy an environmental agent to keep pests from eating your vegetables. You find two products, read their names and wonder whether you should make a purchase. One label reads “plant protection product,” the other “Pesticide.” What information will help you decide whether to buy either?

**DIRECTIONS:**

Have a member of your group read the information on the grid to the group. As a group, discuss and answer each question in the grid. (You may use a dictionary for help.) Write your thoughts in the spaces provided. When you have finished the grid, discuss questions 1–5 and answer each question in the space provided. Finally, individually, write a paragraph telling whether you would buy either product. Also tell what helped you make that decision.

<table>
<thead>
<tr>
<th>PRODUCT DESCRIPTION</th>
<th>“PLANT PROTECTION PRODUCT”</th>
<th>“PESTICIDE”</th>
</tr>
</thead>
<tbody>
<tr>
<td>What does this description mean?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Does the description tell how the product works?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>What does the description emphasize about the product?</td>
<td></td>
<td></td>
</tr>
<tr>
<td>How do you feel about using these environmental agents in your home or neighborhood?</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
1. Does either product name tell the user to be careful about using the product? Explain.

2. Which product name might lead to better sales? Explain.

3. Is it possible that a product could be called both a plant protection and pesticide? Explain.

4. If you were interested in using a product and protecting yourself from any possible harm, which name would you prefer? Explain.
S#20 – WHAT’S IN A WORD?

5. Some products that once were called “pesticides” (a chemical used to kill insects, spiders, rodents and other pests) are now called “protection products.”) How do you feel about the name change?

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

Would you make a purchase? What information would help you decide? (Use the information you have already developed to help you explain your decision.) Provide three reasons for your decision.

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________

________________________________________________________________________
SECTION 3 — SUPPORT MATERIALS

MATERIALS AND POSSIBLE SOURCES

POSSIBLE SOURCES

AMERICAN SCIENCE SURPLUS
6901 W. Oklahoma Ave.
Milwaukee, WI 53219
(414) 541-7777
http://www.sciplus.com/

NASCO
(800) 558-9595

NASCO EAST
901 Janesville Avenue
P.O. Box 901
Fort Atkinson, WI 53538
Fax: (920) 563-8296

NASCO WEST
4825 Stoddard Rd.
P.O. Box 3837
Modesto, CA 95352
Fax: (209) 545-1669

SARGENT-WELCH
P.O. Box 5229
Buffalo Grove, IL 60089-5229
(800) 727-4368
Fax: (800) 676-2540
www.sargentwelch.com

SCIENCE KIT & BOREAL
P.O. Box 5003
Tonawanda, NY 14150-5003
(800) 828-7777
Fax: (800) 828-3299
www.sciencekit.com
SECTION 3 — SUPPORT MATERIALS

SOLUTION PREPARATION INSTRUCTIONS

1. DECHLORINATED TAP WATER

This is a student task incorporated in the student experimental module. You will need to treat tap water to remove chlorine since chlorine in the water can kill organisms, as can trace amounts of soap, detergents and some other substances. Use either method to dechlorinate water. Wash and rinse at least five (5) times several one-gallon plastic milk jugs.

Then a few days before beginning the experiment, fill them three-fourths full with tap water and let them stand open to the air for 48 hours so chlorine leaves the water.

2. EXPERIMENTAL TREATMENT SOLUTIONS

The directions that follow are designed for making 500 mL each of “Starting Stock Solution” and “Group Experimental Treatment Solution.” To make 1 liter of each solution use twice the amount of each substance called for in the recipe. For example, to increase the amount of solution from 500 mL to 1000 mL add 100 mL of solution of pesticide to 900 mL of dechlorinated water.

A. 10% STARTING STOCK SOLUTIONS FOR EXPERIMENTAL TREATMENTS

With the class, determine the substances to be tested for their effect on worms. For safety, be sure aprons, goggles and gloves are worn. CAUTION: Consider all substances as toxic.

FOR LIQUID SUBSTANCES

MATERIALS

For each 500 mL of a 10% Starting Stock Solution for pesticides (herbicide, fungicide, insecticide) or other agents (ethanol):

- 500 mL graduated cylinder
- 50 mL graduated cylinder
- 500 mL Erlenmeyer flask and parafilm square, or 500 mL stock bottle w/cap
- Dechlorinated water
- 50 mL of solution of pesticide or other agent
**SECTION 3 — SUPPORT MATERIALS**

**SOLUTION PREPARATION DIRECTIONS FOR STUDENT-DESIGNED EXPERIMENTS**

**PROCEDURE:**

1. Pour into a 500 mL Erlenmeyer flask or stock bottle:
   - 450 mL of dechlorinated water
   - 50 mL of solution of pesticide or other substance

2. Seal or cap and shake well, yet gently

**FROM SOLID SUBSTANCES**

**MATERIALS FOR EACH 500ML OF A 100% AND 10% STARTING STOCK SOLUTIONS:**

<p>| | |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>500 mL graduated cylinder</td>
<td></td>
</tr>
<tr>
<td>Balance</td>
<td></td>
</tr>
<tr>
<td>500 mL Erlenmeyer flask and parafilm square, or 500 mL stock bottle w/cap</td>
<td></td>
</tr>
<tr>
<td>Dechlorinated water</td>
<td></td>
</tr>
<tr>
<td>Weighing paper</td>
<td></td>
</tr>
<tr>
<td>Spatula for weighing</td>
<td></td>
</tr>
<tr>
<td>50 grams of substance to be tested</td>
<td></td>
</tr>
</tbody>
</table>

**PROCEDURE FOR 500ML OF A 10% SOLUTION:**

1. Pour into a 500 mL Erlenmeyer flask or stock bottle:
   - 450 mL of dechlorinated water
   - 50 grams of solid environmental agent

2. Seal or cap and shake well, yet gently, until all of the solid is dissolved.
   (If the solid will not dissolve entirely in the water, mix well several times and let it sit overnight to facilitate dissolution. If solids remain, pour the liquid into a clean 500 mL container and consider this a 100% solution. To make a 10% solution of this dissolved solid, pour into a 500 mL Erlenmeyer flask or stock bottle 450 mL of dechlorinated water and 50 mL of the 100% solution of the solid. Cap and mix.)
SECTION 3 — SUPPORT MATERIALS

SOLUTION PREPARATION DIRECTIONS FOR STUDENT-DESIGNED EXPERIMENTS

B. TREATMENT SOLUTIONS OF 1%, 0.1%, 0.01% & 0.001% CONCENTRATIONS FROM THE 10% STARTING STOCK SOLUTION.

Reminder 1: Be sure all containers, are thoroughly washed and rinsed at least five times so no soap or substance remains on them.

Reminder 2: Dechlorinated tap water is necessary since chlorine in tap water may affect the worms.

Reminder 3: You may need to prepare a second batch of these group stock solutions, depending on the rate at which students use them.

1. THESE DIRECTIONS ARE FOR TEACHER AND STUDENT HELPER PREPARATION OF GROUP SOLUTIONS.
   (Directions for preparation by demonstration by class groups follows in “2” below.)

MATERIALS

- 500 mL graduated cylinder
- 50 mL graduated cylinder
- 500 mL Ehrlenmeyer flask and parafilm square, or 500 mL stock bottle w/cap
- Dechlorinated water
- 500 mL of the 10% starting stock solution

PROCEDURE FOR MAKING 500ML OF 1%, 0.1%, 0.01% & 0.001% SOLUTIONS USING THE 10% STARTING STOCK SOLUTIONS AND DECHLORINATED WATER:

To make a 1% solution, pour into a 500 mL Ehrlenmeyer flask or bottle and mix:

- 450 mL of dechlorinated water
- 50 mL of the 10% starting stock solution of substance

To make a 0.1% solution, pour into a 500 mL Ehrlenmeyer flask or bottle and mix:

- 450 mL of dechlorinated water
- 50 mL of 1% solution of substance
SECTION 3 — SUPPORT MATERIALS

SOLUTION PREPARATION DIRECTIONS FOR STUDENT-DESIGNED EXPERIMENTS

To make a **0.01% solution**, pour into a 500 mL flask or bottle and mix:

- 450 mL of dechlorinated water
- 50 mL of 0.1% solution of substance

To make a **0.001% solution**, pour into a 500 mL flask or bottle and mix:

- 450 mL of dechlorinated water
- 50 mL of 0.01% solution of substance

Be sure to cover flasks with parafilm or put caps on stock bottles.

2. DIRECTIONS FOR STUDENT GROUP DEMONSTRATION OF TREATMENT SOLUTION PREPARATIONS.

*Note to the Teacher:* Making experimental solutions as a class demonstration activity creates interest in and understanding of the science concepts listed below. You may want to practice this activity in an earlier class using colored water so students can see the relationship between dilution and the intensity of the colored water. This process provides an opportunity to discuss the following concepts as they also prepare solutions:

A) Control, solution, diluted, concentrated, “trace” amount  
B) The need for accuracy in measuring  
C) The importance of a clear procedure  
D) The possibility of introducing errors/variables  
E) Ratios or proportions used in making solutions

Remind students of their group assignments and the treatment(s) chosen by their class and group. Depending on the number of groups per class, different concentrations of one or two agents should be investigated. Each group in a class should be assigned to one experimental condition or the control.

Assign two groups the control solution and remaining student groups one concentration each of the particular agent to be tested. Be sure to test a graded series of concentrations of the agent, for example a 10%, 1%, 0.1%, 0.01% and 0.001%.

Prepare 10%, 1%, 0.1%, 0.01 and 0.001% group experimental treatment and control solutions according to student-done class demonstration guided by the teacher.

Have class materials at the front lab bench with graduated cylinders labeled “Dechlorinated Water” and “Agent.”
SECTION 3 — SUPPORT MATERIALS

SOLUTION PREPARATION DIRECTIONS FOR STUDENT-DESIGNED EXPERIMENTS

CLASS MATERIALS – AT FRONT LAB BENCH (PER CLASS OF 10 GROUPS):

- 10 liters dechlorinated water
- 2 - 500 mL graduated cylinders
- 2 - 100 mL graduated cylinders
- Parafilm (for covering flasks)
- Roll of masking tape
- Permanent marker
- pH testing strips or pH meter
- 10% Starting Stock Solution(s)

GROUP MATERIALS:

- 500 mL Ehrlenmeyer flask or stock bottle, washed and rinsed five times
- Strip of masking tape

PROCEDURE:

STUDENT TEAMS DEMONSTRATE PREPARATION OF GROUP TREATMENT SOLUTION

The first group (1.0% solution group) comes forward with its 500 mL Ehrlenmeyer flask or stock bottle.

1. Measure: 450 mL of dechlorinated water and 50 mL of the 10% Starting Stock Solution of “Agent”
2. Check accuracy of measurements and pour the liquids into a 500 mL flask or bottle.
3. Cover flask with parafilm (bottle with cap) and shake well, but gently, 1/2 minute.
4. Label the container with class, group, treatment solution.
5. Leave your treatment solution on the front lab bench for the next group to use.
6. Rinse “Substance” graduated cylinder with dechlorinated water and shake out the excess water.

The second group (0.1% solution) comes forward with its container.

7. Measure 450 mL of dechlorinated water and 50 mL of the 1% treatment solution of substance.
8. Repeat steps 2 through 6.
SECTION 3 — SUPPORT MATERIALS

SOLUTION PREPARATION DIRECTIONS FOR STUDENT-DESIGNED EXPERIMENTS

The third group (0.01% solution) comes forward with its container

9. To make a 0.01% solution, measure 450 mL of dechlorinated water and 50 mL of 0.1% solution.

10. Repeat steps 2 through 6.

The fourth group (0.001% solution) comes forward with its container

11. To make a 0.001% solution, measure 450 mL of dechlorinated water and 50 mL of 0.01% solution of substance.

12. Repeat steps 2 through 6.

CLOSURE OF THE MATERIALS PREPARATION SESSION

Place prepared group solutions in a designated area. Clean the demonstration area. Have the class summarize what was done to prepare solutions.