



World-Class Support for Science & Math

Living Materials Care and Handling Guide

**For Organisms Used in STC[®], STC/MS[™],
and Other Inquiry-Based Units**

©2004 Carolina Biological Supply Company

Carolina Biological Supply Company

2700 York Rd • Burlington NC 27215
800.334.5551 • www.carolina.com

TABLE OF CONTENTS

INTRODUCTION	4
ARTHROPODS: BUTTERFLIES AND MOTHS	5
Cabbage white butterfly	5
Painted lady butterfly	7
Silkworm	10
Waxworm (greater wax moth)	11
ARTHROPODS: Crustaceans	13
Amphipod	13
Brine shrimp	14
Crayfish	16
<i>Daphnia</i>	18
Fiddler crab	19
ARTHROPODS: Insects	21
Bess beetle	21
Cricket	22
Isopods (pill bug and sow bug)	23
Mealworm (darkling beetle)	25
Milkweed bug	26
Millipede	27
WOWBug™	28
MICROORGANISMS	30
Algae mix	30
<i>Amoeba</i>	31
<i>Blepharisma</i>	32
<i>Euglena</i>	34
<i>Paramecium</i>	35
<i>Spirogyra</i>	37
Vinegar eel	38
<i>Volvox</i>	39
MISCELLANEOUS INVERTEBRATES	40
Annelids (redworm and earthworm)	40
Aquatic snail	41
<i>Hydra</i>	43
<i>Lumbriculus</i> (California blackworm)	44
PLANTS	46
<i>Cabomba</i> , <i>Elodea</i> and <i>Lemna</i> (duckweed)	46
Moss	47
Tree seedling	48
VERTEBRATES	49
Dwarf African frog	49
Fish	51
Internet Resources	53
Aquatic organisms	54
Butterflies and moths	54
Invertebrates	54

ADVISORY ON RELEASING OR DISPOSING OF ORGANISMS..... 54
Living Materials Information Request Form 56

INTRODUCTION

Using live organisms during classroom instruction, especially with inquiry-based science curricula such as STC® and STC/MS™, can greatly enhance and extend student learning. Children have an opportunity to observe, handle, and care for organisms that they probably have never seen or touched before. These first-hand experiences are exciting for youngsters and can translate into improved learning outcomes.

With the use of living organisms in the classroom may come additional instructional concerns and responsibilities for you the teacher. Most organisms have specific habitat requirements such as type of shelter, food and water sources, and temperature range. If these needs aren't met, your organisms will sicken and even die prematurely, which of course will diminish students' learning opportunities. That's why Carolina Biological Supply Company has created this document—to help ensure your use of living materials in the classroom is a successful experience. The care and handling information provided here is similar to or the same as the instructions shipped with your live organisms. If you are using the live organisms as part of a hands-on science kit, these instructions are meant only to supplement, not replace, any directions that may be included in the unit's teacher's guide.

Explanation of Level

Each living organism in this document has been assigned a difficulty level of **Easy**, **Moderate**, or **Advanced**. Admittedly, we have used varying criteria to determine each rating. In some cases (e.g. *Hydra*), the rating of **Advanced** is based on keeping the culture going for an extended period of time. If your intent is to keep the organisms in your classroom for a few days only, then the level of difficulty would change to **Easy**. By way of contrast, the cabbage white butterfly (brassica butterfly), when used with the STC/MS™ hands-on science unit *Organisms—From Macro to Micro*, must be kept alive for an extended length of time in order for students to complete the unit activities. This rates it an **Advanced**. *Daphnia* rates an **Advanced** because of its extreme sensitivity to metal ions that are often found in water.

Technical Support and Questions

We hope your experience using Carolina's live materials is trouble-free. However, if you encounter any problems, please contact us. Direct your living materials question (8 am–5 pm ET, M–F) to one of the following Carolina staff:

- Steve Binkley (800.227.1150, ext. 4304, steve.binkley@carolina.com)
- Tim Woody (800.227.1150, ext. 4381, timothy.woody@carolina.com)

Orders and Replacements

To place an order or request a replacement, contact Carolina at 800.334.5551 (8 am–8 pm ET, M–F), then select Customer Service. If requesting a replacement, please have your order number available.

ARTHROPODS: BUTTERFLIES AND MOTHS

Cabbage white butterfly

(*Pieris rapae*)

Uses: Study of insect characteristics, life cycles, plant-animal relationships

Level: Advanced

Unit(s): STC/MS™: *Organisms—From Macro to Micro*, Grades 6–8

Special requirements: Brassica plants and sugar-water solution



Cabbage white butterflies follow the typical butterfly life cycle: egg, larva (caterpillar), pupa (chrysalis), and adult. Cabbages inhabit Europe, northwest Africa and Asia; they have been introduced to North America and Australia, where they are a serious pest of crops.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER AND INSPECT THE STRIP OF EGGS. The eggs, upon arrival, should hatch 48–72 hours after they are placed on plant

material. Prior to the eggs' arrival, you should have sown at least 10 radish or brassica plants (per unit of eggs) on which to start your larvae. The cabbage white butterfly is especially fond of Wisconsin Fast Plants®, available from Carolina Biological Supply Company. When placing the eggs on the leaves of potted plants, cut the egg strip into 4 to 6 smaller sections and place each section, eggs facing down, on a separate leaf. Immediately after a larva chews its way out of the egg, it may begin to feed on adjacent eggs on the strip. For that reason, enough extra eggs are included in each unit to ensure the 5 or 6 successful hatches expected.

Important: If you neglected to prepare radish or brassica seedlings on which to hatch the eggs, you need to get some cabbage leaves right away. If possible, use organically grown produce, free of pesticides. To remove possible pesticide residue from grocery store produce and ensure the health of the larvae, be sure to remove the outer leaves and discard. Rinse the inner leaves thoroughly with cool water and pat dry with a clean paper towel. Tear off a few leaves and place them in 3 or 4 plastic containers, such as 12-oz deli cups. Cut your egg strip into smaller pieces and place each piece, eggs facing down, on one or more cabbage leaves. The larvae develop more slowly and are more difficult to see on the cabbage leaves than on potted plants. Allow 5 or 6 days before expecting to see the larvae.

Note: Cabbage white butterfly larvae are extremely small and translucent in color when they first hatch, so they are difficult to see. If you think your eggs have not hatched and there are no larvae, please do the following before calling Carolina for a replacement. Look for signs of something chewing on your plants and/or for *frass* (larvae excrement). The larvae are usually found on the underside of the leaf. Using a small hand magnifier may be helpful.

The tiny larvae crawl off the egg strip and immediately feed on the plant material. Larvae molt 2 to 3 times in the first week. When molting, larvae seek a dry site, weave a fine carpet of silk,

attach to it, lie quietly, then crack and crawl out of their exoskeleton and quickly pump up their new exoskeleton before it rigidifies.

The larvae's excrement, frass, dries to fine, dark-black granules, which may be used as fertilizer for additional plants. When using cabbage leaves, you will need to carefully remove the cabbage and larvae from the dish occasionally to clean out the frass. Otherwise, the larvae's frass might encourage the growth of mold on the food plants. Also, remove any old, dry leaves and replace them with fresh ones whenever needed.

Once the larvae you placed on potted plants have eaten them down, you can gently remove the larvae and either place them on more plants you have prepared, or place them on any available brassicas, such as cabbage, brussels sprouts, or broccoli. These should be set up in plastic containers. Be sure to remove the outer leaves of the brassica to eliminate any pesticide residue. Use a brush to remove the larvae; they should not be physically handled prior to the fourth instar (approximately 12–15 days old) because they are tiny and can be easily injured.

After 18–20 days, the larvae will begin to prepare to pupate. Transition from larva to adult occurs during the pupal stage within the chrysalis and is known as *metamorphosis*. When preparing for pupal formation, a larva will weave a strong carpet of silk on the plant or container. (Larvae have silk glands in both the anterior and posterior ends.) The larva then weaves a silk belt around its middle and attaches its rear end to the carpet. The exoskeleton of the pupa is initially soft. Allow the exoskeleton to harden for several hours, and then gently remove the pupa from its attachment site and use double-stick tape to attach it to a piece of paper. Hang the paper inside your butterfly box or cage. Approximately 6–7 days after pupa formation, a butterfly should emerge. Newly emerged butterflies will hang quietly to allow their wings to expand and harden. Wings usually take 15 minutes to expand, then another several hours to harden.

The butterflies should be kept at room temperature (22° to 24° C, 72° to 76° F). If you want them to mate and lay eggs, provide them with a light in or near their cage for 18 hours a day. If you are just keeping the butterflies to observe and enjoy, normal room lighting is sufficient.

Sugar, water, minerals, and other nutrients found in floral nectar are the primary food source of adult cabbage white butterflies. Water and energy from sugar are essential. Without them, adult butterflies die in a few days.

The mouthparts of adult butterflies function solely to take in water and dissolved nutrients. The butterfly *proboscis* is an elongated feeding tube that can be rolled up or extended through the use of specialized muscles. The length of the extended proboscis varies greatly from species to species, depending on the particular flowers the butterflies are adapted to feed on.

If you have planted Wisconsin Fast Plants for your butterflies and they are flowering, the butterflies will quickly forage for nectar from the flowers. If the flowers are not open or are past flowering, or if you did not use Wisconsin Fast Plants, then an artificial feeder can be made according to the following directions:

Obtain an empty plastic film canister. Clean it with a mild solution of household bleach (5%) in water and then rinse it **thoroughly**. Fill it with warm water. Add 1 teaspoon of sugar, 2–3

drops of honey, and 2 drops of yellow food coloring. Stir well. Punch a hole in the lid of the canister, large enough to easily insert a couple of white felt wicks. The wicks should be approximately 1/4 inch wide by 2 inches long. Dip the felt pieces into the sugar-water solution and then squeeze out the excess water. Insert the felt into the hole in the lid, leaving approximately 3/4 inch extending out the top of the lid. Place the lid on the canister. Fold the protruding wicks over toward the opposite sides of the canister, so the wicks create the 2 “petals” of a flower. The sugar/water solution should be replaced every other day. With proper nutrition, the adult butterflies generally live for 1 or 2 weeks, sometimes as long as 3 weeks.

Follow all other care and handling instructions in your Teacher’s Guide.

FAQs

Q. What are brassica plants?

A. Brassicas are members of a large family of flowering plants, many of which are grown for food. Examples include radish, turnip, cabbage, kale, mustard, collards, broccoli, and cauliflower.

Q. I see butterflies in my garden that look like the cabbage white butterfly. Is this possible?

A. Yes. The cabbage white butterfly is found throughout much of North America. It belongs to a family of butterflies that are white, yellow, or orange in color.

Q. The eggs hatched but the larvae died. What caused this?

A. They may have starved. Newly hatched larvae cannot chew through tough plant material as well as older larvae can. Try to start them on young, tender leaves. The humidity also may have been too low. Due to their small size, the larvae can dehydrate rather quickly. If you think humidity is a problem, mist the larvae a few times a day. Use a very fine spray of dechlorinated, room-temperature water. These two problems can be related, since the larvae get moisture from the food they eat. If they are able to feed properly, humidity will seldom be a problem.

For more FAQs, see the painted lady butterfly.

Painted lady butterfly

(Vanessa cardui)

Uses: Life cycle studies, complete metamorphosis, characteristics of insects

Level: Easy

Unit(s): STC®: *The Life Cycle of Butterflies*, Grade 2

Special requirements: Adults (butterflies) require sugar-water solution and flight cage

The painted lady is one of the most common butterflies in the world. It is a migratory species that, because of its wide range, is sometimes called the cosmopolitan butterfly. The 4 distinct stages of its life cycle—egg, larva, chrysalis, and adult—are easily observed.



Larva (caterpillar) and chrysalis (chrysalid): Maintain larvae and cups with food in an area where they receive indirect light; direct sunlight can overheat the cups and kill the larvae. At room temperature larvae will mature in 5 to 10 days. At maturity they climb to the top of the cup and form a pupa, known as a chrysalis (or chrysalid). The chrysalis will be attached to the tissue in the top of the cup. Do not disturb it for at least 1 to 2 days until the chrysalis hardens. Then, open the cup, remove the paper with the chrysalis attached, and tape the paper to the top or side of the butterfly cage so that the chrysalis hangs in a natural position.

Butterfly (adult): Adult butterflies should emerge in 7 to 10 days. They require liquid food. Use the adult feeding system that came in your kit. (You can also make a feeding solution by mixing 2 teaspoons of sugar with 1 cup of water.) Change the feeding solution every 2 to 3 days. The butterflies will live for about 2 weeks.

Note: Butterflies must be prevented from reaching an open container of liquid or they might fall into it and drown.

Eggs: Female butterflies may lay eggs 5 to 7 days after emerging. Look for pinhead-sized, mint-green dots. Fertile eggs hatch in about 3 days. To grow an additional generation of butterflies, you can purchase additional food (catalog no. 14-4040) from Carolina Biological Supply Company. Natural plant foods for the larvae include mallow, thistle, dandelion, and aster.

FAQs

Q. My caterpillars aren't moving. Are they dead?

A. Probably not. Caterpillars often rest for hours at a time, especially just before molting. If you're really in doubt, open the cup and gently touch the caterpillar with the tip of a soft brush. This should cause the caterpillar to move, at least slightly.

Q. Can I take larvae out of the cup?

A. It is best not to open the cup until you are ready to move the chrysalises into the flight cage. There are salts and oils on your hands that can harm the caterpillars if you handle them. Opening the cup might allow bacteria or mold to enter, which could spoil the food.

Q. My caterpillars aren't growing. What's wrong?

A. There are several possibilities; the most common is low temperature at night and over the weekend. If you do not think this is the problem, contact Carolina.

Q. My caterpillars are turning red or pink. What's wrong?

A. The caterpillars have a bacterial infection that will kill them. Contact Carolina for replacements. Discard any sick caterpillars.

Q. Is there enough food for the caterpillars? I'm afraid they will starve.

A. There is plenty of food in the cup to grow all the larvae to maturity.

Q. What do painted lady caterpillars normally eat?

A. In nature, they eat a wide variety of plants including thistle, plantain, hollyhock, mallow, malva, and sunflower. Most of these plants are considered weeds.

Q. My butterflies just came out and there's red liquid all over. Is it blood? Are my butterflies OK?

A. Your butterflies are fine. The red liquid is not blood. It's *meconium*—liquid waste mixed with extra pigment left over from wing formation. It can make a mess on the floor of the cage. You may want to place a paper towel on the cage floor before moving the chrysalises. This will help keep the cage cleaner. (By the way, insect blood is clear, not red.)

Q. Some of the butterflies have crumpled wings. What can I do?

A. When the butterflies first emerge, their wings are crumpled and soft. The butterfly pumps blood into its wings to expand them. If the humidity is too low, the wings may dry and harden before they can expand. Try misting the inside of the cage to raise the humidity level. Also, if a chrysalis falls to the bottom of the cage, the butterfly will not be able to expand its wings properly when it emerges. Place a drop of nontoxic glue on the tip of the chrysalis's abdomen and glue it back on the paper. Once the glue dries, hang the paper in the cage.

Q. What is a chrysalis?

A. Insects that have complete metamorphosis have 4 stages in their life cycle: egg, caterpillar (larva), pupa, and adult. The chrysalis is a pupa with a hard outer shell. Many moths produce a cocoon. A cocoon is a silk case that contains a pupa.

Q. Can my butterflies feed from flowers?

A. Yes, nectar from flowers is a natural food. Place a pot of petunias, marigolds, or other flowering plant in the butterfly cage. It may help to sprinkle the plant with sugar water. Once the butterflies discover the flowers, you'll probably see them feeding.

Q. Can I use cut flowers?

A. Yes, but only if you put them in a vase with a narrow neck so the butterflies cannot fall into the water and drown.

Q. I used some plants as you suggested. Now I see some pinhead-sized green things on the leaves. Are these butterfly eggs? What do I do?

A. Congratulations! They are eggs. Not everyone is so lucky. The eggs will hatch after 3 to 5 days. If it is warm weather, you can try growing them on collected leaves. Tender young leaves are best. Try members of the aster family, which includes thistles, daisies, coneflowers, black-eyed susans, etc. Also try malva and hollyhock. You can purchase more of the special painted lady food from Carolina (catalog no. 14-4040).

Q. I only see 2 pair of legs on each butterfly. What happened to the other pair of legs?

A. The painted lady does not use its first pair of legs for walking. Instead, these legs are feathery and wrap around the butterfly's neck almost like a scarf.

Q. How can I tell the male from the female?

A. Female butterflies have a larger, more rounded abdomen, which is caused by the egg mass. The abdomen of a male, when viewed from the top, has fairly straight sides. From the same view, the sides of the female's abdomen curve outward.

Silkworm

(*Bombyx mori*)

Uses: Study of the characteristics of insects, life cycle, complete metamorphosis

Level: Moderate

Special requirements: Mulberry leaves or artificial diet, warmth, darkness



The silk spun by silkworms has been woven into some of the most beautiful fabric in the world. Silkworm production began over 5,000 years ago in China. The beauty and luster of silk made it the exclusive possession of royalty in China. The Chinese closely guarded the secrets of silk production for thousands of years. Not until the 11th century did European traders manage to steal a few eggs and carry them to Europe to begin rearing

silkworms. Today, Japan is the leading producer of silk because its climate is conducive to the commercial growing of mulberry trees (the sole food of silkworms). Each cocoon a silkworm spins is composed of a single thread, and it takes nearly 3,000 cocoons to make one pound of silk.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER AND INSPECT YOUR SHIPMENT. Silkworm larvae feed on the leaves of mulberry trees and will not eat any other natural food. Mulberry leaves must be available to the larvae immediately after they hatch from their eggs. Mulberry trees (*Morus* sp.) occur widely across the United States, and the young, tender leaves needed for the larvae are available in early spring.

Older leaves are too tough for the first through third instar larvae. (An *instar* is the stage between molts. Silkworms have 5 larval instars.) The fourth and fifth instars can eat the tougher leaves. The leaves must be free of pesticides. As an alternative, an artificial diet is now available for the growth of silkworms.

The first 3 larval instars do best at an optimal temperature of 29° C (84° F). In any case, the temperature should not drop below 20° C (68° F) for young larvae. The fourth and fifth instars are more temperature tolerant and can be raised at room temperature. Check often to ensure all larvae have food and that the leaves are moist and fresh.

FAQs

Q. My silkworms have stopped eating. What's wrong?

A. They may be preparing to molt. At this time they become somewhat brighter in color and stop eating. As the larvae grow larger, they consume more food and need more room to prevent overcrowding. If they aren't preparing to molt and you are raising them on mulberry leaves, replace the leaves with fresh ones. If you are raising them on artificial diet, the diet may have become too dry. Replace it with fresh diet.

Q. The larvae grew large, but now seem smaller. What's happening?

A. During the sixth to eighth day of the fifth instar, the bodies of the silkworms shrink slightly and become somewhat transparent. This indicates the silkworms are ready to spin their cocoons. Construct cocoon nests according to the directions in your Teacher's Guide. Store the cocoon nests in a dark place at 25° C (77° F). A mature larva needs about 3 days to spin its cocoon. Once the cocoon is completed, it takes another 2 to 3 days for the larva to pupate.

Q. My silkworm eggs haven't hatched.

A. Allow enough time. It takes 2 to 3 weeks for the eggs to hatch. Mist the eggs lightly to keep them from drying. Check the eggs frequently for hatchlings. The eggs turn dark just before hatching.

Q. My silkworms hatched, but they don't seem to be eating.

A. If you are growing them on mulberry leaves, the leaves may be too old, or you may not have mulberry leaves. If you are growing them on artificial diet, the diet may have become too dry. Replace it with fresh diet.

Q. The silkworms were growing, but now they have stopped and are turning black and dying. What is wrong?

A. The silkworms are overcrowded. As they grow larger, they need more space.

Q. The moths have emerged, but they can't fly. What's wrong?

A. Nothing. Adult silkworm moths cannot fly. Silk growers do not want their moths flying away, so they have selected and bred moths that cannot fly. Moths begin emerging about 2 weeks after cocoon formation. They tend to emerge in the early morning. The females are larger and less active than the males, which flutter their wings and crawl about in search of females.

Q. What should I feed the moths?

A. Moths do not feed. In fact, they don't have mouthparts. Their job is to reproduce.

Waxworm (greater wax moth)

(Galleria mellonella)

Uses: Study of the characteristics of insects, life cycle

Level: Easy

Special requirements: Warm temperatures

(Photo by H.A. Turney)



The waxworm is commonly found in beehives, where the larva consumes the wax honeycomb and thus poses a serious problem for beekeepers. However, the waxworm has several advantages for use in the classroom—it is extremely easy to maintain and demonstrates complete metamorphosis.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER AND INSPECT YOUR SHIPMENT. The moth is easily reared, hardy, odorless, and can be handled safely. The culture we ship is self-contained, and the moth will complete its life cycle in the vessel in which it is shipped. Follow the instructions included with your shipment. The mated female moth deposits small, white, slightly oblong eggs in masses in cracks away from the light. Newly hatched larvae immediately begin to burrow into the wax. Young larvae are white, about 1 mm long, and very active. Mature larvae are about 25 mm long, golden gray or brown, and move much more slowly. When ready to pupate, the larvae spin cocoons. The moth is about 2 cm long with a wingspread of 2.5 to 3 cm. The male is slightly smaller than the female and has scalloped wing edges.

FAQs

Q. What is the length of the life cycle?

A. This depends greatly on temperature. At temperatures of 29° to 34° C (84° to 92° F), development from egg to adult takes 4 to 6 weeks.

Q. What are the stages of the life cycle?

A. In the temperature range given above: eggs, 5 to 8 days; larva, 14 days; pupa, 8 days; moth, 2 weeks. At lower temperatures, these stages can be extended by weeks or even months.

Q. Can the waxworms be refrigerated?

A. Yes, although there is little reason to do so. Refrigeration will bring the waxworm's development to a virtual standstill.

Q. What can I do with the waxworms after we finish studying them?

A. Do not release waxworms because they are detrimental to honeybees, which are important pollinators. The culture is easily maintained by following the instructions that we provide, so you could pass the culture along to another teacher. If this is not possible, destroy the waxworms by thoroughly freezing the culture.

ARTHROPODS: CRUSTACEANS

Amphipod

(*Gammarus* sp.)

Uses: Study of structure and function, behavior, consumers in aquatic habitats

Level: Moderate

Special requirements: Spring water



Although they are not familiar to most people, amphipods are common in aquatic ecosystems. They are crustaceans and are related to lobsters, crayfish, crabs, and shrimp. *Gammarus* is the most common freshwater genus, and amphipods are sometimes called *Gammarus* shrimp or freshwater shrimp. Amphipod means “2 legs” and refers to the presence of 2 different types of thoracic legs. There are 2 pairs of shorter legs toward the front, and 3 pairs of longer legs to the rear. Amphipods are usually flattened from side to side and have large compound eyes on the sides of the head. They often swim on their sides, which gives them the common name of “sideswimmer.” They are primarily scavengers of plant and animal material, although they may attack injured or stressed animals. They avoid light and usually hide when not swimming about or mating.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER AND INSPECT YOUR SHIPMENT. Remove the lid from the shipping jar and allow it to just sit on the jar, but DO NOT AERATE THE CULTURE WITH A PIPETTE OR ANY OTHER DEVICE. Keep the jar in a cool area (21° C, 69° F) out of direct sunlight. Amphipods are best maintained in a glass or plastic container (e.g., 20-liter or larger aquarium) filled with spring water or bottled water. Do not use city tap water or distilled water because the organisms are sensitive to metal ions, which are usually present in at least trace amounts. Amphipods feed on algae, as well as on bacteria and yeast.

Prepare as follows a yeast suspension as food for amphipods. You can use a rinsed 2-liter soft drink bottle and spring water. Stir in enough baker’s yeast to make the water milky, and store the suspension in a refrigerator. Always agitate the water before use to resuspend the yeast, and feed the amphipods a few drops each day. An alternative is to crush 3 to 4 grains of dry baker’s yeast on clean paper and dust this on the surface of the culture. Avoid overfeeding. Hard-boiled egg yolk or powdered egg yolk can be used in the same amounts as yeast to encourage the growth of bacteria. However, if bacteria overgrow, they can kill the animals. The water in the culture should not become clouded. Each week draw off and discard about 1/4 of the culture water and replace it with new spring water. Follow the instructions in your Teacher’s Guide.

*If you keep your amphipod culture long enough, water will evaporate out of the culture jar and will need to be replaced. Tap water is not suitable, so use spring water or filtered pond water. We do *not* recommend using the “spring water” available at most grocery or drug stores; it may contain trace amounts of metal ions that can kill the amphipods. The best source of replacement water is pond water that has been poured through cheesecloth, or boiled and allowed to cool to room temperature.

FAQs

Q. How can I tell male from female amphipods?

A. During breeding season, this is easy. Female amphipods have a dark brood pouch on the underside of their abdomens.

Q. Can I keep amphipods in an aquarium with fish?

A. The fish will eat them. In fact, many aquarium hobbyists culture amphipods to feed their fish.

Q. Can I refrigerate amphipods?

A. We do not recommend refrigeration of these organisms. Plan on using the culture as soon as possible after its arrival.

Brine shrimp

(*Artemia salina*)

Uses: Study structure and function, behavior, consumers in aquatic habitats

Level: Moderate

Special requirements: Saltwater, light



development and young hatch from the cysts.

Brine shrimp are small crustaceans found in salt lakes and brine pools. Under ideal conditions, female brine shrimp can produce eggs that hatch soon after emerging from their ovisacs to produce live young. When environmental conditions become less than ideal, such as during periods of high salinity or food shortage, the female responds by producing dormant cysts, which are encased embryos that cease development until conditions are again favorable. The dormant cysts can remain viable for years if kept in a dry, oxygen-free environment. Thus the widely sold brine shrimp “eggs” are actually brine shrimp cysts. Once the cysts are incubated in saltwater, the embryos quickly resume their

Hatching brine shrimp eggs: Two-liter soft drink bottles with their tops cut off are good for hatching brine shrimp. Use aged, conditioned tap water throughout. In one liter of water, dissolve 2 tablespoonfuls of noniodized salt. The exact amount is not critical. Synthetic sea salt is best, but rock salt also works. This is enough saltwater for hatching 1/4 tablespoon to one level tablespoon of brine shrimp eggs. Constant light is needed for hatching, so you need a lamp. Drop in a coarse-bubbling air stone or other bubbler to provided needed circulation and oxygen.

Under these conditions and at temperatures of 26° to 28° C (80° to 82° F), the eggs hatch in 24 hours. Lower temperatures result in longer hatching times. Do not exceed 30° C (86° F), or the young may be damaged. When hatching is complete (not all cysts hatch), remove the air stone and direct the light to the middle of the bottle. The shells of the old cysts float, unhatched cysts

settle to the bottom, and the young shrimp (nauplii) concentrate in the light. You can skim off most of the old shells and discard them.

To study brine shrimp, collect them with a clean pipette or pour the culture through a net to capture masses. If you plan to feed the brine shrimp to fish, rinse them first under running water to remove the salts and metabolites (materials that might harm the fish). If you plan to grow the brine shrimp to adulthood, transfer them to a clean bottle (or other nonmetallic container that is free of soap residue) of saltwater.

To prepare a yeast suspension as food for the shrimp, make up a salt solution as before. Stir in enough baker's yeast to make the water milky, and store the yeast suspension in a refrigerator. Always agitate the water before use to resuspend the yeast. Feed your shrimp a few drops of the yeast suspension each day. An alternative is to crush 3 to 4 grains of dry baker's yeast on clean paper and dust this on the surface of the brine shrimp culture. Avoid overfeeding. The water in the culture should not become clouded. Each week draw off and discard about 1/4 of the culture water and replace it with new saltwater. As the shrimp grow, you may have to set up additional containers to avoid overcrowding.

FAQs

Q. I had many cysts that didn't hatch. What should I do with them?

A. Most of these cysts are slow hatchers. Mix up a new bottle of saltwater and add them to it. They should hatch on the second try.

Q. How long does it take brine shrimp to reach adulthood?

A. Under good conditions, nauplii (newly hatched brine shrimp) grow rapidly, reaching adulthood in 8 days. The adults average about 8 mm in body length, but can be double that length.

Q. How can I tell if my brine shrimp are healthy?

A. Shine a flashlight into the bottle. If they do not concentrate in the light, they are not healthy. If you have a microscope, examine the digestive tract, which is a straight tube running the length of the shrimp's body. It should be full of food.

Q. Can I refrigerate brine shrimp?

A. They can be refrigerated (not frozen) for several days. Feed them several hours before refrigeration.

Q. Why are my brine shrimp dying?

A. They could be overcrowded. Divide the culture. There could be insufficient aeration, or you could be using a wooden air stone or other air stone that produces a fine "mist" of bubbles. These small bubbles can clog the shrimp's feeding system and starve them.

Crayfish

(*Orconectes immunis*, *Procambarus clarkii*)

Uses: Study of structure and function, behavior

Level: Moderate

Special requirements: Aquaria with aged and conditioned water



Crayfish are crustaceans, relatives of lobsters, crabs, and shrimp, most of which are marine animals. Crayfish are the only large crustaceans commonly found in freshwater ponds and streams. They are omnivorous, but primarily vegetarian, favoring rotting leaves and plant detritus. Opportunistic feeders, they eat just about anything including vegetables, fish food, fish, manure of any type, plants, wood, and meat. They are also cannibalistic, particularly if they are overcrowded or not enough food is available.

Aquaria: Follow the directions in your Teacher's Guide for setting up the aquaria. Treat all water for holding tanks and aquaria with the water conditioner that is included in your kit. The water level in the tank does not need to be more than 15 cm deep. Give some thought to where you will keep the aquaria. It is best to select an area where the temperature is fairly constant, between 18° and 25° C (64° and 77° F). An aquarium needs diffuse light and should not be placed in direct sunlight or unusually bright artificial light.

Remember that crayfish and other aquarium animals are cold-blooded. An aquarium in direct sunlight may have a temperature as much as 5° C above the temperature of the room. At night such an aquarium cools rapidly, stressing the fish and leaving them vulnerable to disease. To the aquarium you can add stones with a rough surface, a small piece of hollow log, or a short length of PVC pipe for the crayfish to climb on or hide in. There should be slightly more hiding places than the number of crayfish in the aquarium. Adding water plants also provides hiding places, and the crayfish will eat the plants, too. Water snails can be added to help keep the tank clean and prevent the growth of algae. Crayfish also eat snails.

Acclimating crayfish: IMPORTANT! IMMEDIATELY ACCLIMATE CRAYFISH TO HOLDING PAILS OR HABITATS—DO NOT LEAVE THEM IN SHIPPING BAGS! Allow 50 to 60 minutes to acclimate your crayfish. Extra time spent now will head off serious problems, so resist the temptation to rush. Read the relevant sections of your Teacher's Guide. Your aquaria should be ready with conditioned, room temperature water. Avoid the 3 deadly stresses: temperature shock, pH stress, and toxin buildup.

Immediately upon arrival of the crayfish, open the top of the bag and let in some fresh air. Then either float the bag in the holding tank or simply allow it to come to room temperature (propped in a container to prevent spilling). After 20 to 30 minutes, remove and discard about 1/4 of the water from the bag and replace it with water from the holding tank or with other prepared water (aged or dechlorinated) from the same supply as that in the holding tank. Wait 15 minutes and repeat the above step. After another 15 minutes, carefully remove the crayfish with a net and

place them into the holding tank. The water in the shipping bag has accumulated wastes and should be discarded.

For more detailed instructions on crayfish care and water preparation, see your Teacher's Guide.

FAQs

Q. Our crayfish stay hidden and never come out to eat. What is wrong?

A. Crayfish are nocturnal. Feeding behavior is mostly controlled by the amount of light filtering through the water, so usually the greatest periods of activity occur shortly before dawn and just after dusk. Water temperature also plays a role in activity. At the temperature extremes, feeding rate will decrease as will the metabolic rate, which will result in reduced growth.

Q. How much should we feed our crayfish?

A. It is important not to overfeed the crayfish. Feed sparingly and remove uneaten food after a few minutes to avoid fouling the water.

Q. How many crayfish can be kept together?

A. Crayfish are very susceptible to overcrowding. Ideally, there should be no more than 3 small crayfish per cubic meter of water. In small tanks, you should only have one or 2 crayfish.

Q. Can I refrigerate the crayfish?

A. No. However, they do better if placed in a cool area of your classroom.

Q. How can I keep my crayfish healthy?

A. The period following the introduction of crayfish into an aquarium is critical. Acclimate your crayfish properly, and they will be off to a good start. Maintain the aquarium to keep them healthy. Once each month remove about 1/4 of the aquarium water and replace it with fresh water that has been aged and treated with water conditioner. This dilutes any toxic buildup in the aquarium and keeps the pH and ionic balance of the water within a normal range. Never replace more than 1/4 of the water at any one time. Avoid sudden changes to the aquarium. Stressed crayfish often become sick crayfish, so keep their environment stable and fresh.

Q. All the crayfish died soon after we put them in the aquaria. What went wrong?

A. Did you properly acclimate the crayfish? Did you use the water conditioner? Some city water systems now use chlorinators that are not removed by aging the tap water. In those cities, the water conditioner must be used to dechlorinate the water. If you acclimated the crayfish properly and used the water conditioner, your tap water may contain metal ions that are toxic to crayfish. This is especially likely if the water pipes at your school are less than 3 years old or if there have been recent, major repairs to the plumbing. You may have to use water from another source or bottled water. Finally, the crayfish may have been overstressed in shipping. Request a replacement from Carolina.

Daphnia

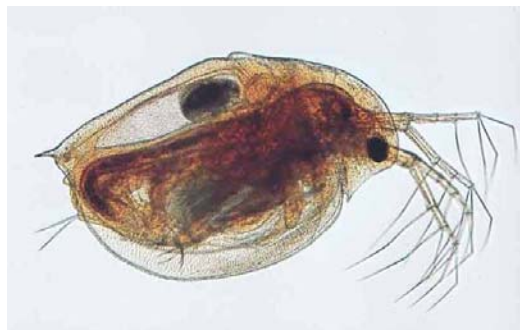
(*Daphnia pulex*)

Uses: Study of structure and function, behavior, consumers in aquatic habitats, pulsation of heart

Level: Advanced

Unit(s): STC/MS™: *Organisms—From Macro to Micro*, Grades 6–8

Special requirements: Spring water or filtered pond water*



The *Daphnia* genus is a common small crustacean of freshwater ponds and lakes. *Daphnia* swim with their antennae and feed with their legs. They are characterized by a “jerky” method of movement, which occurs because the large antennae are used as oars, causing the body to jump forward as the antennae snap backwards. This “jumping” movement gives *Daphnia* the common name of “water flea.” A filter feeder, the thoracic legs act as sieves for filtering

algae, bacteria, and small particles of debris from the water. Food is transferred to the mouth where it is ground by the mandibles and moved through the gut for digestion. A *Daphnia* is highly transparent—a stereomicroscope allows you to see its heart beat.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER AND INSPECT YOUR SHIPMENT. Remove the lid from the shipping jar and allow the lid to just sit on top of the jar. DO NOT AERATE THE CULTURE WITH A PIPETTE OR ANY OTHER DEVICE. Keep the jar in a cool area (21° C, 69° F) out of direct sunlight.

Daphnia feed on algae and are sometimes used to clear up “green water” in an aquarium. They also feed on bacteria and yeast. Prepare as follows a yeast suspension as food for *Daphnia*. You can use a rinsed 2-liter soft drink bottle and spring water. Stir in enough baker’s yeast to make the water milky, and store the suspension in a refrigerator. Always agitate the water before use to resuspend the yeast, using a few drops each day. An alternative is to crush 3 to 4 grains of dry baker’s yeast on clean paper and dust this on the surface of the culture. Avoid overfeeding. Hard-boiled egg yolk or powdered egg yolk can be used in the same amounts as yeast to encourage the growth of bacteria. However, if bacteria overgrow, they can kill the animals.

The water in the culture should not become clouded. Each week draw off and discard about 1/4 of the culture water and replace it with new spring water. Do not use city tap water or distilled water because *Daphnia* are extremely sensitive to metal ions, which are usually present in at least trace amounts. In fact, *Daphnia* are so sensitive to contaminants that they are used to monitor the water quality of streams and lakes.

Follow any additional instructions in your Teacher’s Guide.

*If you keep your *Daphnia* culture long enough, water will evaporate out of the culture jar and will need to be replaced. Tap water is not suitable, so use spring water or filtered pond water. We do *not* recommend using the “spring water” available at most grocery or drug stores; it may contain trace amounts of metal ions that can kill the

Daphnia. The best source of replacement water is pond water that has been poured through cheesecloth, or boiled and allowed to cool to room temperature.

FAQs

Q. I poured my *Daphnia* culture into an aquarium, and they all floated. What went wrong?

A. *Daphnia* have an expanded carapace, a part of the exoskeleton that encloses the sides of the thorax and abdomen, with a space between the carapace and the body. When you poured the daphnia, air became trapped in this space, causing them to float to the surface where they were trapped. Never aerate daphnia or put an air stone in an aquarium or holding tank that contains them. Add daphnia to an aquarium by gently submerging the open jar and “pouring” them underwater.

Q. How can I tell male from female *Daphnia*?

A. During breeding season, this is easy. Female daphnia have a dark egg sac inside the carapace and on the back.

Q. Can I keep daphnia in an aquarium with fish?

A. The fish will eat them. In fact, many aquarium hobbyists culture daphnia to feed their fish.

Q. Can I refrigerate daphnia?

A. We do not recommend refrigeration of these organisms. Plan on using the culture as soon as possible after its arrival.

Fiddler crab

(*Uca pugilator*, *Uca pugnax*)

Uses: Study of the characteristics of animals, semiterrestrial habitat, behavior

Level: Moderate to advanced

Unit(s): STC®: *Animal Studies*, Grade 4

Special requirements: Access to brackish water



Fiddler crabs are small burrowing crustaceans. Functioning as scavengers, they live along the shores of beaches, salt marshes, and estuaries in tropical and temperate zones. The crabs supplied by Carolina Biological Supply Company are the freshwater type and are easy to maintain in the classroom.

Fiddler crabs are easily sexed. The female has 2 small claws, which are used for feeding. The male has one large claw and one small claw; he uses his large claw to threaten other fiddlers and to attract females during the mating season. The waving motion of the large claw has been compared to the movements a musician makes while playing the fiddle, hence the name “fiddler crab.” Occasionally, the male loses his large claw during a fight. The missing claw will be replaced

during the next molt; however, the large claw will be on the opposite side. This explains why some fiddlers have the large claw on the right while others have it on the left.

Terraria: Follow the directions in your Teacher’s Guide for setting up the specialized terraria needed by fiddler crabs. Give some thought to where you will keep the terraria. Remember that direct sunlight may overheat and kill the crabs, so place the terraria in an area of indirect light. Maintain at room temperature (20° to 25° C, 68° to 77° F). The crabs will be more active at the warmer temperatures.

Acclimating and holding fiddler crabs: Refer to Appendix A in the *Animal Studies* Teacher’s Guide and prepare the holding pails prior to the arrival of the crabs. **IMPORTANT! IMMEDIATELY RELEASE CRABS TO HOLDING PAILS OR HABITATS—DO NOT LEAVE THEM IN THE SHIPPING BAGS!** Open the bags slightly to pour out and discard the shipping water. Carefully release half the crabs into each holding pail. Put one pinch (4 to 5 pieces) of dried plankton in the water, and one pinch on the sand. Do not overfeed the crabs. Punch air holes in the holding pail lids and then cover the pails. Provide a safe environment (i.e., room temperature, indirect light), and do not handle the crabs until you are ready to use them.

Care: Keep the water bowl half full at all times, and change it when it becomes dirty. The sand should not be deeper than the height of the water bowl. Keep the lid on the terrarium after the crabs are placed inside.

FAQs

Q. How do I keep my fiddler crabs healthy?

A. The most important things to do are to keep them at a consistent temperature and to keep the water in the bowl changed on a regular basis.

Q. Will the fiddler crabs mate?

A. You may observe courtship behavior in which the male waves his large claw to attract the female. Usually the male must have a burrow before the female will mate with him. There is not enough sand in the terrarium for the male to excavate a burrow, although he may try.

Q. How long do fiddler crabs live?

A. In the wild, no one knows. In captivity they can live for a school year with good care.

Q. Can I keep 2 or more males in a terrarium?

A. Probably. Male fiddler crabs may wrestle, push, and shove each other, especially in the presence of a female. These matches rarely result in injury, but a losing crab may be stressed.

Q. A crab has molted. What do I do?

A. In most cases, no additional care is needed. Do not remove the molted skin. Over the next several days the crab will eat it, providing needed calcium. The crab will be fragile for several days after a molt. If another crab is bothering it, remove it for a few days.

ARTHROPODS: INSECTS

Bess beetle

(*Odontotaenius disjunctus*)

Uses: Study of the characteristics of animals, characteristics of invertebrates, consumers in terrestrial food webs and chains

Level: Easy

Unit(s): STC®: *Organisms*, Grade 1

Special requirements: Moist terrarium, rotten hardwood



The bess beetle is a large, shiny, black beetle 1 1/4 to 1 3/8 inches long, sometimes called the patent leather beetle. Its body is divided into 3 distinct parts: head, thorax, and abdomen. The bess beetle has 3 pairs of legs, a pair of antennae, and strong jaws that protrude from the front of its head. It eats microorganisms found in wood, chewing through damp, rotting wood to create tunnels inside the log where it lives.

The larvae (which are white) and adults (which feed the larvae with prechewed wood) share the tunnels. In this sense, they are social insects. However, they are not as organized as termites, ants, wasps, or bees, which divide jobs among the group. Adults and larvae stridulate, meaning they produce sounds. Adults stridulate by rubbing a rough area under their wings against a similar area on the top of their abdomens. Larvae stridulate by rubbing the tip of the third leg against the joint of the second leg. Although adult beetles have 2 pairs of wings, they rarely, if ever, fly. They are easily maintained if kept in damp (not soggy) wood, and they prefer darkness.

Terraria: Follow the directions in your Teacher's Guide for setting up the terraria. Think about where you will keep the terraria. Remember that direct sunlight may overheat and kill the beetles, so place the terrarium in an area of indirect light. Maintain at room temperature (20° to 22° C, 68° to 72° F). Bess beetles need humidity, so the terrarium soil should be damp. There should be decaying organic material (wood, leaves, compost, etc.) in the terrarium. Mist the habitat to keep it humid, but if mold becomes a problem, increase the ventilation or reduce the amount of misting.

Care: Other than humidity, bess beetles need little care. If they eat all the rotten wood shipped with them, collect additional rotten wood (preferably hardwood) locally.

FAQs

Q. Can bess beetles bite?

A. Bess beetles are harmless. Although their jaws are strong, they are adapted for chewing rotten wood, not human skin. Bess beetles can be safely handled as described in your Teacher's Guide. Notice the hooks on the beetles' legs. These can cling so tightly to some fabrics, especially the

yarn of sweaters, that the beetle may be injured while being pulled off. For this reason, don't allow beetles to crawl on clothing.

Q. What is that squeaking sound?

A. That's called stridulation, as explained above. Since the beetles are somewhat social, the sounds may be communications among colony members.

Q. How long do bess beetles live?

A. Bess beetles can live as adults for up to 1 1/2 years, unusually long for a beetle. They also undergo complete metamorphosis, so their entire life cycle (egg, larva, pupa, and adult) can extend beyond 2 years. The beetles you receive are of unknown age, but they will probably live for at least 6 months.

Q. Will my bess beetles reproduce?

A. This is unlikely. They must have a very stable environment, like that provided by a large rotting log, for reproduction.

Q. Since they eat wood, aren't bess beetles destructive?

A. No, they feed on the fungi and bacteria in decaying wood. Although they help break down wood that is already damaged, they do not attack undamaged wood.

Cricket

(Acheta domesticus)

Uses: Study the characteristics of animals, characteristics of invertebrates, consumers in terrestrial food webs and chains, reproduction, life cycle, incomplete metamorphosis

Level: Easy

Unit(s): STC®: *Ecosystems*, Grade 5

Special requirements: Terrarium



The house cricket, kept as a pet in many parts of the world, is a useful classroom insect for demonstrating incomplete metamorphosis. In nature, a female cricket lays eggs in moist sand or soil in the late fall. To do this, she inserts her long ovipositor into the sand and deposits 50–100 eggs, one at a time. The eggs are small, banana-shaped, and yellowish or white. The eggs overwinter, and the nymphs emerge in the spring. At room temperature in the classroom, the eggs hatch in 2 to 3 weeks. Newly hatched nymphs are about the same size as the egg and difficult to see. Nymphs are similar in appearance to adults but lack wings. They undergo several molts, finally emerging as winged adults.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER, REMOVE THE CUPS, AND INSPECT THEM. Because of the short life span of adults, we ship cricket nymphs that are about 1/4 inch long. Crickets are easy to keep. They are

primarily herbivorous but accept a variety of foods. Fresh apples, pears, and lettuce are foods that provide moisture, while dry dog food provides high protein and cuts down on cannibalism. Crickets tolerate higher temperatures than do many other classroom animals.

FAQs

Q. Can crickets bite?

A. No, crickets are harmless.

Q. Why do crickets chirp?

A. Adult males chirp to attract females.

Q. Our crickets developed into adults and then died. What happened?

A. The life span of an adult cricket is about 2 weeks, so your crickets probably lived out their normal life span. Did you observe the females laying eggs? If so, watch for the hatching of young.

Q. Young crickets have hatched. How long will it take them to grow into adult crickets?

A. Their growth is strongly influenced by temperature. At normal classroom temperature it will take about 60 to 90 days.

Q. Our crickets died without producing any young. What went wrong?

A. Assuming that you had male and female crickets together, your room temperature may have been too low. Crickets are much more likely to reproduce at higher room temperatures. Also, crickets may cannibalize their own eggs or young if food is scarce.

Isopods (pill bug and sow bug)

(Armadillidium vulgare, Porcellio laevis)

Uses: Characteristics of animals, characteristics of invertebrates, consumers in terrestrial food webs and chains, behavior

Level: Easy

Unit(s): STC®: *Organisms*, Grade 1; *Ecosystems*, Grade 5

Special requirements: Moist terrarium



(Pictured: [top image] pill bugs; [bottom image] sow bug)

Terrestrial isopods are land-dwelling crustaceans commonly known as sow bugs or pill bugs. They have many other common names: potato bugs, wood lice (no relation to body lice), and roly-polies are just a few. Related to lobsters, crabs, and shrimp, terrestrial isopods breathe with gills and require a humid environment for survival. They inhabit cool, damp places under rocks, rotting wood, and other decaying vegetation. Although similar in size, color, and life cycle, pill bugs and sow bugs are



different. When threatened, pill bugs can curl up into a tight ball for protection, while sow bugs either run or remain perfectly still, appearing to be dead.

Terraria: Follow the directions in your Teacher's Guide for setting up the terraria. Give some thought to where you will keep the terraria. Remember that direct sunlight may overheat and kill the isopods, so place the terrarium in an area of indirect light. Maintain at room temperature (20° to 22° C, 68° to 72°

F). Isopods need humidity, so the terrarium soil should be damp, and there should be decaying organic material (wood, leaves, compost, etc.) in the terrarium. Mist the habitat to keep it humid, but if mold becomes a problem, increase the ventilation or reduce the amount of water used to mist the habitat.

Acclimating and holding isopods: IMMEDIATELY OPEN THE SHIPPING CONTAINER OF ISOPODS AND CHECK THEIR CONDITION. They are shipped in a damp paper towel to provide the humidity they require. After examining the isopods, close the container and prepare a holding pail. Punch air holes in the lid, and place a cup of moist potting soil in the bottom of the pail. Put a slice of raw potato or apple on the soil. Mist the inside of the pail with room-temperature water. Empty the contents of the shipping container onto the bottom of the pail. Gently unfold the crumpled, moist paper towel and shake off as many of the isopods as possible. If you are unable to dislodge all the isopods, leave the paper towel in a loose tangle in the pail, and replace the lid. Leave the pail in a darkened area for at least an hour. Most of the isopods should leave the paper towel and move down to the damp soil. Use a camel hair brush to remove any that remain. Mist the inside of the pail again and replace the lid. The isopods can be held like this for several hours or overnight until they can be transferred into their habitat.

Care: Other than humidity, isopods need little care. Feed them fish food flakes, leaf litter, and pieces of raw potatoes, apples, or carrots. Immediately remove food that becomes moldy.

FAQs

Q. Our pill bugs disappeared. Where are they?

A. Isopods avoid light. They are probably under some leaf litter, or they have burrowed into the soil in the terrarium. Place a piece of raw potato or apple on the surface of the soil and leave it overnight. Gently lift the slice of raw vegetable and you will probably find the isopods underneath. You are more likely to observe the isopods if you keep the terrarium in a dimly lit or dark area of your room.

Q. Our pill bugs died while we were observing them with magnifiers. What happened?

A. Without proper humidity, isopods die because their gills dry quickly. To observe isopods with a hand lens, place them in a shallow cup with some damp soil or paper towel. When observations are complete, return the isopods to the terrarium as soon as possible.

Q. We are finding lots of tiny bugs in our terrarium. They look a bit like the pill bugs, but are smaller. What are they?

A. You have provided a good habitat for the isopods and they are reproducing. The female carries up to 200 eggs in a brood pouch located under her thorax. The young hatch in the pouch and stay there for about 3 weeks. They resemble the adults, except for their smaller size and paler color, and will molt 4 to 5 times as they grow.

Mealworm (darkling beetle)

(Tenebrio obscurus)

Uses: Study of the characteristics of insects, life cycle, complete metamorphosis, consumers in land habitats, behavior

Level: Moderate

Special requirements: Moisture, protection from light



Mealworms are the larvae of the darkling beetle (*Tenebrio*), which undergoes complete metamorphosis: egg, larva (mealworm), pupa, and adult (beetle). A female beetle lays several hundred eggs. You are unlikely to see them because they are sticky and rapidly become coated with debris. Eggs hatch about 2 weeks after they are laid, but it may take a few weeks before the larvae are large enough to notice. The length of the larval stage varies considerably with

temperature and food availability. Under ideal conditions, the larvae grow rapidly until they are about 25 mm long, and then they pupate. The adults emerge 2 to 3 weeks after pupation.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER, REMOVE THE CUPS, AND INSPECT THE ORGANISMS. You will receive either mealworms (larvae) or adults (darkling beetles) according to which hands-on unit you are using. Once you have verified that the shipment is OK, the insects are ready to use. To culture mealworms, put 5 to 10 cm of bran meal in the bottom of a plastic pan. Other grains, such as oatmeal, wheat germ, and whole-wheat flour, can also be used. Add 25 to 50 beetles and place a small slice of potato on top of the meal for moisture. A thin layer of shredded paper can be added for the beetles to crawl on. Mealworms may also be cultured in chicken mash, which has been spread on layers of moistened burlap. Use a small slice of potato as before. You can substitute a slice of apple or a banana peel if you prefer.

After 3 to 5 months, check the culture regularly for adults and pupae. These can be removed and used to set up new cultures. The beetles have wings but seldom fly. Still, it is a good idea to cover the culture container with a screen or perforated lid. As the name “darkling beetle” implies, these insects prefer the dark, so keep them in a cabinet or other darkened location. Mealworms (all stages) do well at normal room temperatures but will develop fastest at temperatures in the 25° to 30° C (77° to 86° F) range.

FAQs

Q. Do these insects bite?

A. No. They are completely harmless to people.

Q. Can mealworms be refrigerated?

A. We recommend that all live materials be used as soon as possible; however, all stages of *Tenebrio* can be refrigerated. Do not freeze.

Q. Are mealworms pests?

A. Mealworms can damage stored grain. They are generally beneficial because they feed on organic matter not usually eaten by other animals. They are also food for other animals. Some people raise mealworms as live food for pet reptiles or wild birds.

Q. How can I tell the male beetles from the females?

A. That isn't practical, as it involves dissection of specimens and detailed knowledge. Male and female beetles probably recognize each other with special scents or pheromones.

Milkweed bug

(Oncopeltus fasciatus)

Uses: Study of the characteristics of insects, life cycle, incomplete metamorphosis

Level: Easy

Special requirements: Raw, cracked sunflower seeds



Although the term “bug” is often incorrectly applied to all insects, only those insects in the order Hemiptera are true bugs. The milkweed bug is a true bug, and it is ideal for studying the structure and life cycle of this important group of insects. Its development from egg to adult is an example of incomplete metamorphosis. The young nymphs closely resemble the adults, but they do not have wings or reproductive organs. In nature the milkweed bug feeds on several species of milkweeds.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER AND INSPECT YOUR SHIPMENT. At 29° C (84° F) the egg stage lasts 4 days, so hatching should begin soon. The color of the egg gradually changes from yellow to deep orange as it nears hatching. The newly emerged nymph is about the size of a pinhead and is bright orange. The nymph grows by a series of molts. At 29° C it takes about a month for the nymph to become an adult, which lives for about an additional month. Raw, cracked sunflower seeds serve as food for both adults and nymphs. Follow the instructions in your Teacher's Guide to set up and maintain your cultures of milkweed bugs.

FAQs

Q. How often should I change the sunflower seeds?

A. The nymphs and adults do not completely eat the seeds. Replace the seeds when they become shrunken or dirty.

Q. How can I tell male from female?

A. This is easy in the adult bugs. The ventral side of the fourth abdominal segment (counting from the thorax) bears a black band in the male and two prominent black spots in the female.

Q. How can I tell nymphs from adults?

A. The young nymphs closely resemble the adults but do not have wings. As is typical of insects that undergo incomplete metamorphosis, the wing pads begin to appear in the early stages, gradually increase in size at each molt, and become prominent in the last nymph stage before the molt to the adult stage.

Q. When will the bugs produce eggs?

A. Mating takes place 5 to 12 days after the last molt for females and 2 to 3 days for males. Egg laying begins 1 to 15 days after mating and peaks at about 20 days.

Millipede

(*Orthoporus texicolons*)

Uses: Study of the characteristics of animals, characteristics of invertebrates, consumers in terrestrial food webs and chains

Level: Easy

Unit(s): STC®: *Organisms*, Grade 1; *Animal Studies*, Grade 4

Special requirements: Moist terrarium



Millipedes are also known as thousand-legged worms. Although none actually have 1,000 legs, large millipedes certainly have several hundred legs, 2 per body segment. The legs work in groups to produce locomotion; thus, waves of motion travel down the rows of legs as the animal crawls. This allows a millipede to exert a lot of force as it pushes its way through leaf litter or loose soil.

The harder the animal has to push, the greater the number of legs involved in generating the push. They are herbivores, eating mostly decomposing vegetable material.

Millipedes are secretive animals with few defenses, so they hide under any available material, although some forage in the open at night. Primarily they depend on their hard exoskeletons for protection. When disturbed, many curl up, protecting their soft undersides. Millipedes do not bite or sting, but many have repugnatorial glands, which secrete a liquid that can be irritating to the skin. The millipedes provided by Carolina Biological Supply Company have a very mild

secretion, but even so, when handling them, avoid touching your eyes or mouth, and wash your hands afterwards.

Terraria: Follow the directions in your Teacher’s Guide for setting up the terraria. Give some thought to where you will keep the terraria. Remember that direct sunlight may overheat and kill the millipedes, so place the terrarium in an area of indirect light. Maintain at room temperature (20° to 22° C, 68° to 72° F). Millipedes need humidity, so the terrarium soil should be damp, and there should be decaying organic material (wood, leaves, compost, etc.) in the terrarium. Mist the habitat to keep it humid, but if mold becomes a problem, increase the ventilation or reduce the amount of water used to mist the habitat.

Care: Other than humidity, millipedes need little care. Feed them fish food flakes, leaf litter, and pieces of raw potatoes, apples, lettuce, mushrooms, or carrots. Even though millipedes feed on rotting vegetation in nature, remove any old food from the habitat if mold begins to develop. In case there are any baby millipedes, check old food before discarding it—the babies often attach themselves to it.

FAQs

Q. Our millipedes disappeared. Where are they?

A. Millipedes avoid light. They are probably under some leaf litter, or they have burrowed into the soil in the terrarium. You are more likely to observe the millipedes if you keep the terrarium in a dimly lit or dark area of your room.

Q. How do I keep my millipedes healthy?

A. Remember that millipedes need humidity. Regularly spray the habitat with room temperature conditioned water. Chitin is essential to millipedes for molting and growth of a new exoskeleton. Oak leaves are rich sources of this nutrient, so be sure to add leaf litter to the habitat. Millipedes thrive under somewhat unkempt conditions, so don’t be too fussy about keeping their terrarium clean. Given this preference, millipedes are themselves clean animals. They spend a lot of time cleaning their antennae, exoskeleton, and legs.

Q. Our millipedes have tiny bugs on them. What are they?

A. They are probably mites, which are symbiotic with the millipedes and do not harm them. The mites clean up bits of rotting materials that are discarded or missed by the millipedes.

WOWBug™

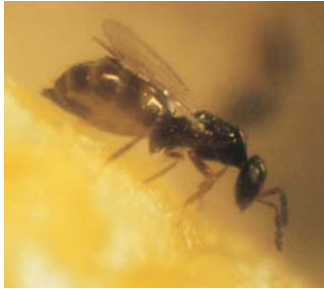
(Melittobia digitata)

Uses: Example of a parasite, study of behavior

Level: Moderate

Unit(s): STC/MS™: *Organisms—From Macro to Micro*, Grades 6–8

Special requirements: None



(Pictured: [top image] female; [bottom image] male)

WOWBugs are small, harmless, parasitic wasps unable to sting humans. Found around the globe, they are easy to handle, and no expensive equipment is needed to study them. All stages of the life cycle are readily visible: their eggs are attached to the outside of the host insect's body, and their pupae are naked, lacking any cocoon to cover them. The pupae of the WOWBugs are cream-colored and approximately 1/8 inch long. A prolific reproduction rate helps make WOWBugs ideal for classroom use.



Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER AND INSPECT YOUR SHIPMENT. Your culture of WOWBugs was shipped while the insects were in the pupal stage. Some adults may have emerged while in transit. If the majority of the WOWBugs are still in the pupal stage, simply wait several days and adults will emerge. There is no need to feed the adult WOWBugs.

Maintain the WOWBugs at room temperature. At normal room temperatures of 22° to 30° C (72° to 86° F), the entire WOWBug life cycle (egg, larva, pupa, adult) takes 18–25 days. While in the pupal stage, the WOWBug does not move around or eat. WOWBugs have never been observed feeding during the adult stage, although some evidence suggests that males cannibalize one another, and it is possible that females may feed on juices from their host organism.

For long-term storage, cultures are best refrigerated when WOWBugs are in the prepupal stage (fully fed larvae). This most closely mimics the stage at which they spend the winter outdoors. Should it be absolutely necessary to refrigerate adults, keep them cold for as short a time as possible, and store them in the butter compartment on the refrigerator door where the temperature is slightly warmer. Don't be alarmed if they don't move right away when you take them out. It sometimes takes up to an hour for them to warm up enough to begin to behave normally.

FAQs

Q. What are the dark, pill-like things that I received?

A. These are puparia, cases that enclose fly pupae. WOWBug larvae are parasites of fly pupae. After the WOWBug larvae grow to full size, they stop feeding and become pupae themselves. They emerge as tiny adult wasps. The adult wasps should begin emerging about the time you receive your shipment.

Q. Will adult flies emerge from the puparia?

A. This is possible but unlikely. The fly pupae have been so heavily parasitized by WOWBug larvae that they probably will be unable to emerge. If they do, they can be killed by freezing.

MICROORGANISMS

Algae mix

(*Ankistrodesmus falcatus*, *Haematococcus droebakensis*, *Selenastrum capricornutum*)

Uses: Study of the characteristics of microorganisms, producers in aquatic food webs and chains

Level: Easy

Unit(s): STC®: *Ecosystems*, Grade 5

Special requirements: Aquarium, light



Algae is a catchall word that includes organisms that live in water (mostly) and that use the energy of sunlight to manufacture sugars, i.e., the process of photosynthesis. This makes algae important producers in many aquatic and marine habitats. Indeed, algae have been called “the grass of many waters.” Algae are grouped according to the types of photosynthetic pigments they produce. Thus, there are blue-green, red, and brown algae. The mixed algae used in STC® are members of the green algae group; that is, they have the same photosynthetic pigments (chlorophylls a and b) found

in green plants.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER, REMOVE THE JARS OF ALGAE, AND INSPECT THEM. The water in the jars of mixed algae should be pale green. Once you have verified that the shipment is OK, loosen the lids on the jars. These organisms need light but not heat. In fact, a cool room temperature is better for them. Indirect natural light is good, but not direct sunlight. Fluorescent light, either cool white or full spectrum, is good. Avoid incandescent lamps, as they can overheat the water and kill your algae. Plan to use your algae as soon after receipt as possible. Note: No additional water is needed. Do not dilute the algae.

FAQs

Q. How long can I keep my algae cultures before using them?

A. Algae are shipped to be in prime condition when you receive them. The sooner you use them, the better. At most, plan to keep algae cultures no longer than 2 to 3 days before using them.

Q. Will the algae last longer if I place the jars in a refrigerator?

A. We don't recommend refrigeration or rapid temperature changes.

Q. Can my students look at the mixed algae with a microscope?

A. Yes, but they need magnification ranging from 70 to 100x to see much. The algae in this mix are unicellular (single cells) or form small colonies of cells, and each cell is quite small, so it may take some searching to find any algae cells.

Amoeba

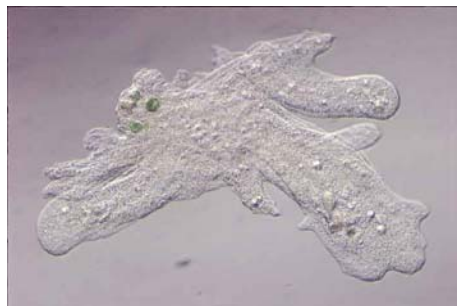
(*Amoeba proteus*)

Uses: Study the characteristics of microorganisms, diversity of microorganisms, cell structure and function, cell movement

Level: Moderate

Unit(s): STC/MS™: *Organisms—From Macro to Micro*, Grades 6–8

Special requirements: Spring water or filtered pond water*



The body of an *Amoeba* and other protozoans consists of a single cell, but it is a mistake to think of these organisms as “simple.” Without benefit of multicellular tissues or organs, many protozoa achieve structural complexity that rivals that of some multicellular animals. For this reason, some biologists prefer to think of protozoan organization as “acellular” rather than “unicellular.” *Amoebae* are found in freshwater locations like ponds and streams, typically on decaying vegetation.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER AND REMOVE THE CULTURE JAR. Carefully open the jar and aerate the culture using the pipette supplied (use only new or biologically clean pipettes). To aerate the culture, place the tip of the pipette in the upper layer of liquid in the culture jar and gently press the pipette bulb. This will blow air bubbles into the liquid. Remove the pipette tip from the liquid and allow the pipette to refill with air, then blow more air bubbles into the liquid. Do this several times.

Allow 15 to 20 minutes after aeration for the animals to settle, and then inspect the contents using a microscope at low light level. If you receive more than one culture in your shipment, use a different pipette for each culture, and write the name of the organism on the pipette to avoid cross-contamination. Warn students against contaminating or accidentally killing a culture. Students should use the provided pipettes.

Never place the culture in a refrigerator or in direct sunlight. It should be kept cool (20° to 22° C, 68° to 72° F) with the lid placed lightly over the jar. Some protozoan cultures react noticeably to environmental changes. If the temperature suddenly drops, *Amoebae* become sluggish. Animals that are actively feeding or undergoing mitosis tend to ball up. This does not mean they are dead or dying; dead protozoans disintegrate, leaving no visible remains. The organisms may arrive partially covered with metabolic debris, but this is also normal. Careful examination of the cultures using a microscope should reassure you that they are still healthy. In an undisturbed culture, most *Amoebae* will settle to the bottom of the container, where they can be seen with a stereomicroscope.

Using a stereomicroscope and a clean pipette, you can easily pick up a single specimen (or group of specimens) for a slide. If a stereomicroscope is not available, very carefully draw into a pipette a small amount of material from the bottom of the jar. One drop should contain more than

enough organisms for one good slide mount. Follow any other instructions in your Teacher's Guide.

We provide all protozoan cultures with an ample food source, some of which may not have been consumed. This is not contamination. After several weeks, however, all food may have been consumed. If you wish to feed your *Amoebae*, place 3 or 4 grains of previously boiled wheat seed in the culture jar. Add spring water or pond water as needed. The water in the culture should not become clouded. Each week draw off and discard about 1/4 of the culture water and replace it with new spring water.

*If you keep your protozoan culture long enough, water will evaporate out of the culture jar and will need to be replaced. Tap water is not suitable, so use spring water or filtered pond water. We do *not* recommend using the "spring water" available at most grocery or drug stores; it may contain trace amounts of metal ions that can kill the protozoans. The best source of replacement water is pond water that has been filtered (poured through cheesecloth), or boiled and allowed to cool to room temperature.

FAQs

Q. We haven't been able to find any *Amoebae*. Where are they?

A. Because they are small, colorless, slow moving, and do not have a fixed shape, *Amoebae* can be difficult to find. Allow the culture jar to remain undisturbed for 15 to 20 minutes. Most of the *Amoebae* will settle to the bottom and begin to crawl about. Place the culture jar on the stage of a stereomicroscope at 20 to 40x and focus on the inside bottom of the jar. You may have to watch for several seconds, but you should begin to see *Amoebae* as they slowly move about. Once you see one, it will be easier to find others.

Q. Are these *Amoebae* dangerous?

A. No. There are parasitic *Amoebae* that can cause illness, but these *Amoebae* are free-living and harmless.

Blepharisma

(*Blepharisma americanum*)

Uses: Study of the characteristics of microorganisms

Level: Easy

Unit(s): STC®: *Microworlds*, Grade 5

Special requirements: None



Blepharisma is a member of the Protista group of organisms. Its body consists of a single cell, which is animal-like in that it ingests food particles and moves about. *Blepharisma* moves by the coordinated beating of cilia, small hairlike structures that cover much of its surface. *Blepharisma* is pale pink.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER, REMOVE THE CULTURE JARS, AND INSPECT THEM. Once you have verified that the shipment is OK, loosen the lids on the jars. Aerate the cultures using the pipettes supplied. To aerate, place the tip of a pipette into the culture water and squeeze the bulb, bubbling air into the water. Withdraw the pipette and release the bulb, allowing it to refill with air. Repeat about 4 times. This replaces oxygen, which may have been depleted during shipment. IMPORTANT: Use a different pipette for each culture, and mark each pipette so that it will always be used with that culture only. This is especially important if you also have vinegar eels, because EVEN A TRACE OF VINEGAR CAN KILL THE *BLEPHARISMA* AND MOST OTHER MICROORGANISMS, INCLUDING ALGAE. After aerating the cultures, place the lids on the jars, but do not tighten them. Store the cultures at room temperature (*Blepharisma* does better at lower room temperatures), out of direct sunlight. A dim corner of the room works well. Use the cultures as soon as possible.

*If you keep your *Blepharisma* culture long enough, water will evaporate out of the culture jar and will need to be replaced. Tap water is not suitable, so use spring water or filtered pond water. We do *not* recommend using the “spring water” available at most grocery or drug stores; it may contain trace amounts of metal ions that can kill the *Blepharisma*. The best source of replacement water is pond water that has been poured through cheesecloth, or boiled and allowed to cool to room temperature.

FAQs

Q. How long can I keep my cultures before using them?

A. If possible, use them within 2 to 3 days of receipt. The longer you delay, the more likely the cultures will go bad, be knocked over, etc.

Q. Will the cultures last longer if I place the jars in a refrigerator?

A. We don't recommend refrigeration or rapid temperature changes. Refrigeration may kill the organisms.

Q. Can I make my cultures last longer?

A. *Blepharisma* is easy to culture. You can find instructions in Appendix E of the *Microworlds* Teacher's Guide. This would make a good project for your students.

Q. My students aren't finding any *Blepharisma*. What can I do?

A. The culture may have been agitated, scattering the *Blepharisma*. Look at the culture after it has sat undisturbed for at least 15 minutes. You should see some debris and fuzzy material near the bottom of the jar. Bacteria, which the *Blepharisma* feed on, are found in the fuzzy material. *Blepharisma* concentrate in this area to feed. To sample this area, squeeze the bulb of a pipette (BE CERTAIN THIS IS NOT THE PIPETTE USED WITH ANOTHER CULTURE, SUCH AS THE VINEGAR EELS!) and insert the tip vertically into the culture. When the tip is just above or actually in the fuzzy material, release the bulb. Lift the pipette vertically out of the culture. As long as you do not bubble air into the culture, squirt the water back into the culture, or use the pipette to stir the culture, the *Blepharisma* remain concentrated and easy to sample.

Euglena

(*Euglena gracilis*)

Uses: Study the characteristics of microorganisms, diversity of microorganisms, cell movement

Level: Easy

Unit(s): STC/MS™: *Organisms—From Macro to Micro*, Grades 6–8

Special requirements: Spring water or filtered pond water*



Euglena exhibits characteristics of both plants and animals. Like a plant, *Euglena* contains chlorophyll, allowing it to make its own food through photosynthesis. When light is not available and it cannot photosynthesize, *Euglena* resembles other animals—it gains nutrients by absorbing them across its cell membrane.

Euglenae use a whiplike flagellum to move about in a spiral path. You can find this protozoan in a variety of aquatic habitats, both freshwater and marine.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER, REMOVE THE CULTURE JARS, AND INSPECT THEM. Once you have verified that the shipment is OK, loosen the lids on the jars. Aerate the cultures using the pipettes supplied. To aerate, place the tip of a pipette into the culture water and squeeze the bulb, bubbling air into the water. Withdraw the pipette and release the bulb, allowing it to refill with air. Repeat about 4 times. This replaces oxygen, which may have been depleted during shipment.

Allow 15 to 20 minutes after aeration for the animals to settle, and then inspect the contents using a microscope at low light level. If you receive more than one culture in your shipment, use a different pipette for each culture, and write the name of the organism on the pipette to avoid cross-contamination. Warn students against contaminating or accidentally killing a culture. Students should use the provided pipettes.

Never place the culture in a refrigerator or in direct sunlight. *Euglenae* do best in a well-lighted area (artificial light), but out of direct sunlight because high temperatures are harmful. The culture should be kept cool (20° to 22° C, 68° to 72° F) with the lid placed lightly over the jar.

Some protozoan cultures react noticeably to environmental changes. Animals that are actively feeding or undergoing mitosis tend to ball up. Do not be alarmed, for they are not dead or dying; dead protozoans disintegrate, leaving no visible remains. The organisms may arrive partially covered with metabolic debris, but this is also normal. Careful examination of the cultures using a microscope should reassure you that they are still healthy.

Using a stereomicroscope and a clean pipette, you can easily pick up a single specimen (or group of specimens) for a slide. If a stereomicroscope is not available, very carefully draw into a pipette a small amount of material from the bottom of the jar. One drop should contain more than enough organisms for one good slide mount. Follow any other instructions in your Teacher's Guide.

If *Euglenae* are kept in bright light, they will make their own food, eliminating the need for additional feeding by the teacher. The water in the culture should not become clouded. Each week draw off and discard about 1/4 of the culture water and replace it with new spring water.

*If you keep your protozoan culture long enough, water will evaporate out of the culture jar and will need to be replaced. Tap water is not suitable, so use spring water or filtered pond water. We do *not* recommend using the “spring water” available at most grocery or drug stores; it may contain trace amounts of metal ions that can kill the protozoans. The best source of replacement water is pond water that has been filtered (poured through cheesecloth), or boiled and allowed to cool to room temperature.

FAQs

Q. Are *Euglenae* protists or algae?

A. Protists are one-celled (or acellular) organisms with nuclei, so *Euglena* qualifies as a protist. Since it is capable of photosynthesis, it is also an alga. To complicate matters, there are *Euglenae* that do not have chloroplasts and are not photosynthetic.

Q. How long can I keep my cultures before using them?

A. If possible, use them within 2 to 3 days of receipt. The longer you delay, the more likely the cultures will go bad, be knocked over, etc.

Q. Will the cultures last longer if I place the jars in a refrigerator?

A. We don't recommend refrigeration or rapid temperature changes. Refrigeration may kill the organisms.

Q. My students aren't finding any *Euglenae*. What can I do?

A. *Euglenae* are a bit smaller than other protists commonly studied. If your students have previously observed *Paramecia*, they may need to look for something smaller. Let them observe on low power for several seconds. They can use the fine adjustment of the microscope to focus on different levels within the slide. They need to find slowly moving dots or “wiggles.” Once they see one of these, they should center it in the field of vision and switch to a higher power. They may need to refocus slightly using the fine adjustment.

Paramecium

(Paramecium multimicronucleatum)

Uses: Study the characteristics of microorganisms, diversity of microorganisms, cell structure and function, cell movement, consumers in aquatic food webs and chains

Level: Easy

Unit(s): STC/MS™: *Organisms—From Macro to Micro*, Grades 6–8

Special requirements: Spring water*



Paramecium is a small unicellular organism found in freshwater ponds. It swims, rotating slowly, and often changing its direction. To help it move in water,

Paramecium possesses short, hair-like cilia that beat in unison.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER, REMOVE THE CULTURE JARS, AND INSPECT THEM. Once you have verified that the shipment is OK, loosen the lids on the jars. Aerate the cultures using the pipettes supplied. To aerate, place the tip of a pipette into the culture water and squeeze the bulb, bubbling air into the water. Withdraw the pipette and release the bulb, allowing it to refill with air. Repeat about 4 times. This replaces oxygen, which may have been depleted during shipment.

Allow 15 to 20 minutes after aeration for the animals to settle, and then inspect the contents using a microscope at low light level. If you receive more than one culture in your shipment, use a different pipette for each culture, and write the name of the organism on the pipette to avoid cross-contamination. Warn students against contaminating or accidentally killing a culture. Students should use the provided pipettes.

Never place the culture in a refrigerator or in direct sunlight. It should be kept cool (20° to 22° C, 68° to 72° F) with the lid placed lightly over the jar. Some protozoan cultures react noticeably to environmental changes. Animals that are actively feeding or undergoing mitosis tend to ball up. This does not mean they are dead or dying; dead protozoans disintegrate, leaving no visible remains. The organisms may arrive partially covered with metabolic debris, but this is also normal. Careful examination of the cultures using a microscope should reassure you that they are still healthy.

Using a stereomicroscope and a clean pipette, you can easily pick up a single specimen (or group of specimens) for a slide. If a stereomicroscope is not available, very carefully draw into a pipette a small amount of material from the perimeter of the jar. One drop should contain more than enough organisms for one good slide mount. Follow any other instructions in your Teacher's Guide.

We provide all protozoan cultures with an ample food source, some of which may not have been consumed. This is not contamination. After several weeks, however, all food may have been consumed. If you wish to feed your *Paramecia*, place 6 to 8 grains of previously boiled wheat seed in the culture jar. Add spring water or pond water as needed. The water in the culture should not become clouded. Each week draw off and discard about 1/4 of the culture water and replace it with new spring water.

*If you keep your protozoan culture long enough, water will evaporate out of the culture jar and will need to be replaced. Tap water is not suitable, so use spring water or filtered pond water. We do *not* recommend using the "spring water" available at most grocery or drug stores; it may contain trace amounts of metal ions that can kill the protozoans. The best source of replacement water is pond water that has been filtered (poured through cheesecloth), or boiled and allowed to cool to room temperature.

FAQs

Q. How long can I keep my cultures before using them?

A. If possible, use them within 2 to 3 days of receipt. The longer you delay, the more likely the cultures will go bad, be knocked over, etc.

Q. Will the cultures last longer if I place the jars in a refrigerator?

A. We don't recommend refrigeration or rapid temperature changes. Refrigeration may kill the organisms.

Q. Can I make my cultures last longer?

A. *Paramecium* is easy to culture. You can find instructions in the *Carolina Protozoa and Invertebrates Manual*. This would make a good project for your students.

Q. My students aren't finding any *Paramecia*. What can I do?

A. The culture may have been agitated, scattering the *Paramecia*. Look at the culture after it has sat undisturbed for at least 15 minutes. You should see some debris and fuzzy material near the bottom of the jar. Bacteria, which the *Paramecia* feed on, are found in the fuzzy material. *Paramecia* concentrate in this area to feed. To sample this area, squeeze the bulb of a pipette and insert the tip vertically into the culture. When the tip is just above or actually in the fuzzy material, release the bulb. Lift the pipette vertically out of the culture. As long as you do not bubble air into the culture, squirt the water back into the culture, or use the pipette to stir the culture, the *Paramecia* remain concentrated and easy to sample.

Spirogyra

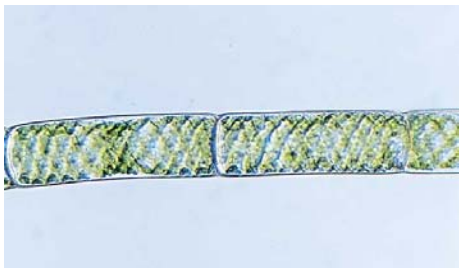
(*Spirogyra grevilleana*)

Uses: Study the characteristics of microorganisms, producers in aquatic food webs and chains

Level: Easy

Unit(s): STC/MS™: *Organisms—From Macro to Micro*, Grades 6–8

Special requirements: Spring water or filtered pond water*



Spirogyra is a freshwater green alga, shaped in chains of hair-like filaments. The green color indicates the presence of chlorophyll, which can be seen as spiral bands within the organism. Chlorophyll enables *Spirogyra* to make its own food through photosynthesis.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER AND

INSPECT YOUR SHIPMENT. Once you have verified that the shipment is OK, loosen the lid on the jar. These organisms need light but not heat. However, most freshwater algae can tolerate a range of temperatures—from 15° to 25° C (60° to 77° F). Indirect natural light is good, but not direct sunlight. Fluorescent light, either cool white or full spectrum, is good. Avoid incandescent lamps, as they can overheat the water and kill your algae. Plan to use your algae as soon after receipt as possible.

*If you keep your algal culture long enough, water will evaporate out of the culture jar and will need to be replaced. Tap water is not suitable, so use spring water or filtered pond water. We do *not* recommend using the “spring water” available at most grocery or drug stores; it may contain trace amounts of metal ions that can kill the algae. The best

source of replacement water is pond water that has been poured through cheesecloth, or boiled and allowed to cool to room temperature.

FAQs

Q. How long can I keep my *Spirogyra* culture before use?

A. Algae, including *Spirogyra*, are shipped to be in prime condition when you receive them. The sooner you use them, the better. At most, plan to keep algae cultures no longer than 2 to 3 days before using them.

Q. Will the *Spirogyra* culture last longer if I place the jars in a refrigerator?

A. We don't recommend refrigeration or rapid temperature changes.

Q. What should I do with the *Spirogyra* culture after we have finished using it?

A. Why not set up a spare 5- to 20-gallon aquarium in a well-lighted area? Add any leftover aquatic organisms to the aquarium and see what happens. Which organisms survive? What are some possible reasons that an organism does or does not survive?

Vinegar eel

(*Turbatrix aceti*)

Uses: Study of the characteristics of microorganisms

Level: Easy

Unit(s): STC®: *Microworlds*, Grade 5

Special requirements: None



The vinegar eel, although microscopic in size, is a true animal. A roundworm, also known as a nematode, it is related to the nematodes that occur in soil. The vinegar eel lives in vinegar, which is highly unusual because vinegar is so acidic that it kills most microorganisms.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER, REMOVE THE CULTURE JARS, AND INSPECT THEM. Once you have

verified that the shipment is OK, loosen the lids on the jars. Aerate the cultures using the pipettes supplied. To aerate, place the tip of a pipette into the culture water and squeeze the bulb, bubbling air into the water. Withdraw the pipette and release the bulb, allowing it to refill with air. Repeat about 4 times. This replaces oxygen, which may have been depleted during shipment. **IMPORTANT:** Use a different pipette for each culture, and mark each pipette so that it will always be used with that culture only. This is especially important for the pipette used with the vinegar eel culture. **EVEN A TRACE OF VINEGAR CAN KILL MOST OTHER MICROORGANISMS, INCLUDING ALGAE.** After aerating the cultures, place the lids on the jars, but do not tighten them. Store the cultures at room temperature out of direct sunlight. A dim corner of the room works well. Use the cultures as soon as possible.

FAQs

Q. How long can I keep my cultures before using them?

A. If possible, use them within 2 to 3 days of receipt. However, the vinegar eel culture is fairly hardy and will probably last for a long time. The longer you delay, the more likely the cultures will go bad, be knocked over, etc.

Q. Will the cultures last longer if I place the jars in a refrigerator?

A. We don't recommend refrigeration or rapid temperature changes. Refrigeration may kill the organisms.

Q. Can I make my cultures last longer?

A. Vinegar eels are easy to culture. You can find instructions in Appendix E of the *Microworlds* Teacher's Guide. This would make a good project for your students.

Q. What if my students can't find the vinegar eels?

A. Place a lamp or flashlight beside the jar so that the light shines through the vinegar from the side. Now observe the vinegar from the side, but don't look directly into the light. You should see "wiggly" areas in the vinegar. The "wiggly" effect is caused by masses of vinegar eels thrashing about, so these are the areas to sample.

Q. Does the vinegar I buy at the store contain vinegar eels? Am I eating these things on my salad?

A. Vinegar purchased at a store has been filtered and sterilized, so relax. Even if you bought "natural" vinegar that had not been filtered, the vinegar eels that might be in it would not harm you.

Volvox

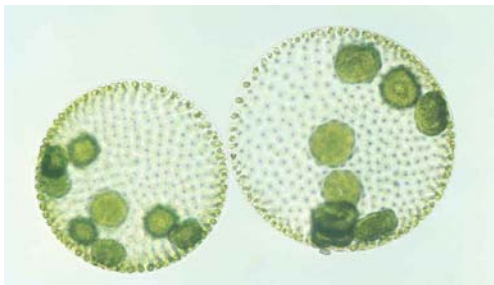
(*Volvox globator*)

Uses: Study the characteristics of microorganisms, producers in aquatic food webs and chains

Level: Easy

Unit(s): STC®: Microworlds, Grade 5; STC/MS™: *Organisms—From Macro to Micro*, Grades 6–8

Special requirements: Spring water or filtered pond water*



Volvox is a member of the green algae group; that is, it has the same photosynthetic pigments (chlorophyll) found in green plants. *Algae* is a catchall word that includes organisms that live in water (mostly) and that use the energy of sunlight to manufacture sugars, i.e., the process of photosynthesis. This makes algae important producers in many aquatic and marine habitats. Indeed, algae have been called “the grass of

many waters.” Algae are grouped according to the types of photosynthetic pigments they produce. Thus, there are blue-green, red, and brown algae.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER, REMOVE THE JAR OF *VOLVOX* AND INSPECT IT. You should be able to see tiny green dots in the water, although a magnifier may be needed for this. Sometimes the best way to see *Volvox* is to remove the lid from the jar and look down from the top. Once you have verified that the shipment is OK, loosen the lid on the jar. These organisms need light but not heat. In fact, a cool room temperature is better for them. Indirect natural light is good, but not direct sunlight. Fluorescent light, either cool white or full spectrum, is good. Avoid incandescent lamps, as they can overheat the water and kill your algae. Plan to use your algae as soon after receipt as possible.

*If you keep your algal culture long enough, water will evaporate out of the culture jar and will need to be replaced. Tap water is not suitable, so use spring water or filtered pond water. We do *not* recommend using the “spring water” available at most grocery or drug stores; it may contain trace amounts of metal ions that can kill the algae. The best source of replacement water is pond water that has been poured through cheesecloth, or boiled and allowed to cool to room temperature.

FAQs

Q. Can I keep the *Volvox* culture longer if I pour off and replace some of the water in the jar?

A. Probably not. Again, use the material as soon as possible.

Q. My students aren’t finding any *Volvox*. What can I do?

A. Try positioning a lamp or flashlight next to the jar so that the light shines through the jar. The *Volvox* colonies should now show up as brightly lit dots. Gently squeeze a pipette bulb and lower the pipette until its mouth is near one of the dots. Release the bulb and the *Volvox* should be pulled into the pipette. Transfer the *Volvox* to a deep-well slide.

MISCELLANEOUS INVERTEBRATES

Annelids (redworm and earthworm)

(*Eisenia foetida*, *Lumbricus terrestris*)

Uses: Study of the characteristics of animals, recycling and change

Level: Easy (redworms) to advanced (earthworms)

Unit(s): STC®: *Soils*, Grade 2

Special requirements: Moisture, protection from light



(Pictured: [top] earthworms compared to [bottom] redworms)

Redworms and earthworms are annelids—segmented worms. They are very similar, but earthworms reach much larger sizes than

redworms do. Both live in soil. Both feed on nonliving organic matter in the soil, but redworms require a much higher organic content than earthworms. Because of this, redworms are often used to help speed composting. Earthworms are important in creating and maintaining soil fertility. Redworms and earthworms avoid sunlight, which can kill them. Earthworms are sometimes called night crawlers from their habit of emerging from the soil at night.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER AND INSPECT YOUR SHIPMENT. Discard any dead worms. The worms can be retained in the shipping materials if you keep them cool and moist. Earthworms require cool temperatures of 15° C (60° F) or lower to do well. This makes it difficult to maintain earthworms for lengthy periods in the classroom. Redworms tolerate higher temperatures, up to 25° C (77° F), which makes them a much better choice for extended classroom use, but anything above 29° C (84° F) can be harmful. See your Teacher's Guide for the care and use of these annelids.

FAQs

Q. How can I tell the difference between male and female earthworms?

A. Each redworm and earthworm is both male and female. In a copulating pair, each worm gives and receives sperm.

Q. Our worms died after a few weeks. What happened?

A. Although they are land animals, redworms and earthworms are dependent on moisture in the material that surrounds them. They breathe through their skins, which must be kept moist at all times. Be certain that they have adequate moisture. Use only aged tap water. In a few cities, aging the tap water does not remove the chlorine compounds, and a special water conditioner must be used in those areas. Keep both redworms and earthworms under darkened conditions because sunlight can kill them. Remember that earthworms need cool temperatures.

Q. Where are earthworms found?

A. Earthworms are found in most soils. They are absent from dry desert areas, and some soils are too acidic for earthworms. Redworms are more common around and in compost and manure piles.

Q. What is the difference between an earthworm and a redworm?

A. Other than average size, the anatomical differences (differences in body structure) are technical and difficult to observe. They do differ in behavior. Redworms live in the upper 25 cm or so of the soil, usually under a thick layer of litter or other organic matter. Earthworms go much deeper and build vertical, more-or-less permanent burrows in which they live.

Aquatic snail

(*Physa gyrin*, *Lymnea columella*, *Planorbis planorbis*, *Planorbella trivoluis*, *Planorbella duryi*, *Gyraulus parvus*)

Uses: Study of the characteristics of animals, characteristics of invertebrates, consumers in aquatic food webs and chains

Level: Easy

Unit(s): STC®: *Organisms*, Grade 1; *Ecosystems*, Grade 5

Special requirements: Aquarium with aged and conditioned water



Like most other mollusks, snails have a shell. In an aquarium, most snails scavenge for food. They eat food left by fish, so they help keep the aquarium clean. Snails may also eat algae that grow on aquarium plants or the sides of the aquarium. If there isn't enough food for them, they may eat aquarium plants.

Aquaria: Follow the directions in your Teacher's Guide for setting up the aquaria. Treat all water for holding tanks and aquaria with the water conditioner included in your kit. Give some thought to where you will keep the aquaria. It is best to select an area where the temperature is fairly constant, between 18° to 25° C (64° to 77° F). An aquarium needs diffuse light and should not be placed in direct sunlight or unusually bright artificial light. Remember that snails and other aquarium animals are cold-blooded. An aquarium in direct sunlight may have a temperature as much as 5° C above the temperature of the room. At night such an aquarium cools rapidly, stressing the snails and leaving them vulnerable to disease.

Acclimating and holding aquatic snails: NOTE: NEWLY ARRIVED SNAILS OFTEN DO NOT SHOW MOVEMENT FOR 2 to 3 DAYS. DO NOT ASSUME THEY ARE DEAD! Immediately open the container of snails—allow at least 30 minutes for snails to reach room temperature. Prepare an appropriate holding pail or habitat. To avoid temperature shock, place snails in room temperature, conditioned tap water. *Elodea* (water plants) may be rinsed and added to the holding pail. If you think a snail is dead, use a pencil to gently pry against the hard flap at the shell opening. If it resists probing or is tightly closed, the animal is alive. Avoid overcrowding the habitat; do not hold more than 15 to 20 snails per 1 to 2 gal of water. Snails may not emerge for 2 to 3 days—this is normal. Immediately change the water if it becomes cloudy or foul smelling, then check for and discard any dead snails.

FAQs

Q. I have a snail that is floating. Does that mean it's dead?

A. Probably not. The snail has collected a gas bubble under its shell. In a few hours or days it will absorb or expel the bubble and sink.

Q. What should I feed my snails?

A. If they are kept in an aquarium with fish and plants, they should not need extra food. They eat any food left by the fish and feed upon other material in the aquarium.

Q. Snails are crawling on the water plants. Are they eating them?

A. Perhaps. They are probably eating algae growing on the plants. However, if there is not enough other food for the snails, they may eat the plants. Also, if the plants are weakened and beginning to die, the snails may eat them.

Q. A snail has retracted into its shell and hasn't moved. Is it dead?

A. Probably not. Snails often do this for a few hours or even for days. As long as the shell is tightly closed, the snail is alive. You can test as described above under "Acclimating and holding pond snails," but it's best not to bother the snails unnecessarily. A snail that hangs limply out of its shell is dead and should be discarded.

Q. All the snails died soon after we put them in the aquaria. What went wrong?

A. Did you properly acclimate the snails? Did you use the water conditioner? Some city water systems now use chlorinators that are not removed by aging the tap water. In those cities, the water conditioner must be used to dechlorinate the water. If you acclimated the snails properly and used the water conditioner, your tap water still may contain metal ions toxic to snails. This is especially likely if the water pipes at your school are less than 3 years old or if there have been recent, major repairs to the plumbing. Snails are more sensitive to metal ions than are most other aquarium animals, including fish. You may have to use water from another source, or bottled water. Finally, the snails may have been overstressed in shipping. Request a replacement from Carolina Biological Supply Company.

Hydra

(*Hydra* sp.)

Uses: Study of structure and function, behavior, consumers in aquatic habitats, regeneration

Level: Moderate

Unit(s): STC/MS™: *Organisms—From Macro to Micro*, Grades 6–8

Special requirements: Spring water



Hydras are freshwater organisms with a tube-like body. A single opening, the mouth, leads into a gastrovascular cavity. The mouth is surrounded by tentacles armed with stinging cells. When a prey organism, such as a small crustacean, brushes against the *Hydra's* tentacles, the *Hydra* first harpoons it with stinging nematocyst threads; it then uses its tentacles to guide the meal into its mouth.

Hydras that live on the undersides of lily pads and other leaves in cool ponds and streams look like tiny pieces of frayed string.

Hydras can move from place to place by "walking" or somersaulting on their tentacles or by sliding along in measuring-worm style.

Sometimes called the "eternal animal," *Hydra* has an amazing ability to regenerate lost body parts. A *Hydra* cut in half will form 2 complete animals within a few days.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER AND INSPECT YOUR SHIPMENT. Leave the lid off the jar for about 30 minutes. If you are not going to use the culture immediately, set the lid loosely on top of the jar, to let air in but keep dust out. Keep the culture in dim light, at a temperature of about 70° F (21° C). The *Hydra* can live for about a week in the water in the culture jar. If you plan to keep the

organisms longer than this, siphon off about 1/3 of the old culture water each day and replace it with fresh spring water.

To feed the *Hydra*, follow the directions provided in your Teacher's Guide.

FAQs

Q. Our *Hydra* appear sick. What's wrong with them?

A. During the sexual phase of their life cycle, *Hydra* put most of their energies into producing sperm and eggs. They contract and appear "sick." This more often happens during the spring season.

Q. Our *Hydra* were doing well, then they all disappeared. What happened?

A. It is difficult to maintain a small culture of *Hydra* for more than a few days. Also, *Hydra* are delicate animals, easily damaged when moved onto a slide for observation. There really isn't much you can do. Plan on using the *Hydra* as quickly as possible after receipt.

***Lumbriculus* (California blackworm)**

(*Lumbriculus variegatus*)

Uses: Study of the characteristics of soft-bodied invertebrates, behavior, regeneration, pulsation of blood vessels

Level: Easy

Unit(s): STC/MS™: *Organisms—From Macro to Micro*, Grades 6–8

Special requirements: Spring water or aged tap water



Lumbriculus resemble miniature earthworms and live in the sediment and silt of freshwater ponds and lakes. A transparent body reveals many of the organism's internal organs.

Care and handling of cultures: IMMEDIATELY UPON RECEIPT, OPEN THE SHIPPING CONTAINER AND INSPECT YOUR SHIPMENT. To maintain your culture of *Lumbriculus*, you will need a deep pan or dish,

spring water (or aged tap water), a plastic pipette, brown paper towel, and sinking fish food. Fill the pan with 2–3 inches of spring water. Transfer the worms into the water with a plastic pipette. Do not handle or transfer worms with forceps or hooks; they are easily injured by these instruments. Add strips of folded brown paper towel to the pan, covering the bottom. This will serve as a substrate of decomposing material for the worms and for numerous microscopic organisms that may inhabit the culture, such as bacteria, rotifers, and ostracods. Folding the paper towel allows *Lumbriculus* to "hide" inside the folds and avoid light, because they prefer darkness.

Lumbriculus will break down quickly (disintegrate) if they are not kept cool. An ideal temperature is about 15° C (60° F). A slightly warmer range (18° to 21° C, 65° to 70° F) is acceptable, but requires that the culture water be changed more frequently—about every 2 to 3 days. Temperatures above 21° C (70° F) will cause organism disintegration.

The sinking fish food is the main food source for the worms. Add 1 or 2 pellets. After a few days, 1 or 2 more pellets can be added if the original pellets have been consumed. Do not overfeed; decomposing food can contaminate the culture, causing a mass die-off of worms. Irregular feedings or weeks of starving will not harm the worms.

Gentle aeration is recommended but not required. To aerate the culture, place the tip of the pipette in the upper layer of liquid in the culture container and gently press the pipette bulb. This will blow air bubbles into the liquid. Remove the pipette tip from the liquid and allow the pipette to refill with air, then blow more air bubbles into the liquid. Do this several times.

Water lost to evaporation is simply replaced by adding spring water. The culture water should be replaced every 2–3 weeks as the paper towel disintegrates and waste residues accumulate. Decant the culture slowly, being careful not to pour out the paper and worms that remain at the bottom. After rinsing the paper and the worms with spring water, refill the pan to the original level and add new pieces of towel. Occasional “harvesting” of surplus worms is advised (for classroom experiments, live fish food, or starting duplicate cultures), and maintenance of at least one duplicate culture is strongly suggested. Under these conditions, worms will reproduce asexually and cultures can be sustained for years.

For additional preparation directions, follow the instructions detailed in Lesson 2 of the Teacher’s Guide.

FAQs

Q. Will the blackworms reproduce?

A. In a culture, blackworms reproduce readily by asexual fragmentation and regeneration. If you are referring to sexual reproduction, this is very unlikely to occur in culture, although it is common in natural populations.

Q. Do blackworms have eyes?

A. No, but they do have sensory cells that can detect shadows. They may rapidly contract when a shadow passes over. This may help them escape from predators. They are also sensitive to pressure changes and vibrations.

PLANTS

***Cabomba*, *Elodea* and *Lemna* (duckweed)**

(*Cabomba caroliniana*, *Elodea densa*, *Elodea canadensis* [also known as *Egeria densa* and *Anacharis densa*], *Lemna minor*)

Uses: Study of the characteristics of plants, characteristics of aquatic plants, producers in aquatic food webs and chains

Level: Easy

Unit(s): STC®: *Organisms*, Grade 1; *Animal Studies*, Grade 4; *Ecosystems*, Grade 5;
STC/MS™: *Organisms—From Macro to Micro*, Grades 6–8

Special requirements: Aquarium

Restrictions: *Elodea densa* (catalog no. 16-2101, 16-2102, and 16-2103) cannot be shipped into SC, WA, or ME. Customers in these restricted states will be shipped *Elodea canadensis*. Canadian orders require a permit from the Canadian government.



(Pictured: [top] *Cabomba*; [bottom] *Elodea*)

Elodea (*Egeria*, *Anacharis*) and *Cabomba* are common aquarium plants. Both can flower and produce seeds, although they more commonly reproduce asexually by stem fragmentation. These plants may root or float free in the water. The most common species are native to the Americas, but they have been introduced worldwide. *Elodea* (*Egeria*) *densa* (commonly called Brazilian *Elodea*), the most common of all aquarium plants, sometimes becomes invasive in streams, lakes, and ponds,

so it should not be released into local waterways. Some states restrict shipments of *Elodea densa*. Carolina Biological Supply Company ships *Elodea canadensis*, which is native to North America, to those states.

Duckweed (*Lemna minor*) flourishes in a variety of environments and can be found in clear water as well as dark, brackish water. This small, floating plant often covers the surface of a pond.

Acclimation of plants: *Elodea* and *Cabomba* are shipped in bundles that are secured with rubber bands and wrapped in wet newspaper. Remove the newspaper and cut the rubber bands. Either rinse the plants in running tap water or swish them about in a pail of aged tap water. This is to clean the plants, so discard the rinse water after you finish. Inspect the plants and discard any that appear unhealthy. Also discard any unwanted snails or other animals that may have hitched a ride on the plants. Place the plants in a holding tank of aged, conditioned tap water until you are ready to use them in class. Don't keep them in the holding tanks too long. If they must be kept for more than one day, replace some of the water (not more than 1/4 of it) with fresh, aged, conditioned water. Also turn the plants over so that any that were covered by others will be exposed to light. The plants need bright light for best growth.

Duckweed does not require a rinse. Simply add the plants to a holding tank of aged, conditioned tap water. Maintain the plants at temperatures between 18° to 25° C (65° and 77° F) for best results. There are no special light requirements.

Follow any additional instructions in your Teacher’s Guide for each of these plants.

FAQs

Q. Why aren’t the plants growing?

A. It can take some time for the plants to adjust to a new home. Fish and other animals in the aquarium release waste products into the water that serve as nutrients for the plants. In a new aquarium, it may take time for these to accumulate, so be patient.

Q. Will the plants form roots?

A. They may or may not. *Elodea* and *Cabomba* often float free in the water. In an aquarium, they are able to absorb nutrients directly from the water, so roots aren’t necessary. If they do form roots, it will probably take some time for them to grow. Duckweed often forms short roots that help absorb nutrients from the water.

Q. Will the fish and snails eat the plants?

A. They may. After all, plants are producers and animals are consumers. If growing conditions are good for the plants, both they and the animals flourish.

Q. Why are my plants dying?

A. There can be many reasons for this. Are the plants getting enough light? Are the aquaria being disturbed too often? The plants must be allowed to “settle in” to grow well. Some nutrient could be missing from the water. A lack of phosphorus and iron may limit the growth of water plants. If possible, add a small amount of water-soluble fertilizer that contains these nutrients. If your water is treated with a water softener (unlikely), that may cause aquarium plants to grow poorly.

Moss

(*Polytrichum commune*, *Dicranum flagellare*, *Hypnum curvifolium*, *Mnium stellare*)

Uses: Study of the characteristics of plants, producers in land food webs and chains

Level: Easy

Unit(s): STC®: *Organisms*, Grade 1; *Animal Studies*, Grade 4

Special requirements: Terrarium, moisture



Mosses are simple plants that occupy many land habitats. A few are aquatic. The mosses used for STC® are found on the floor of forests. Although they are land plants, mosses require moisture and high humidity for healthy growth. Because they grow under trees, they tolerate low light levels. Mosses neither flower nor produce seeds; they reproduce by spores.

Care and handling of plants: IMMEDIATELY UPON RECEIPT OPEN THE SHIPPING CONTAINER AND INSPECT THE PLANTS. The moss plants come sealed in plastic containers. If you can see signs of moisture in the containers, and the moss plants do not appear to be dry, you can leave the moss in the containers until ready to use. If the moss is dry, open the containers and spray the moss with aged room temperature water, and then close the containers.

FAQs

Q. How long can I keep the moss before using it?

A. If possible, use it within 2 to 3 days of receipt. See your Teacher's Guide for instructions for preparing the terraria.

Q. Can the moss be stored in a refrigerator?

A. This isn't recommended because of the danger of freezing. If you do refrigerate the moss, place it in the vegetable bin or another area of the refrigerator with little risk of near-freezing temperatures. This should be an emergency procedure only. It is much better to prepare the terraria and plant the moss as soon as possible.

Q. Are there any special precautions to take with the moss?

A. Follow the planting directions given in your Teacher's Guide. Moss plants require lots of humidity, so you may need to mist the moss daily with aged room temperature water.

Tree seedling

(Araucaria heterophylla)

Uses: Study of the characteristics of plants, producers in land food webs and chains

Level: Easy

Unit(s): STC®: *Organisms*, Grade 1

Special requirements: Terrarium, moisture



The tree seedlings are gymnosperms, which include pines, cypress, hemlocks, firs, and spruce, among others. Many grow into large forest trees. Indeed, the largest trees, sequoias and redwoods, are gymnosperms. Gymnosperms are found from the tropics to the far north. They are of great economic importance, providing us with lumber and wood pulp for paper. Gymnosperms produce seeds in cones, but do not produce flowers.

Care and handling of plants: IMMEDIATELY UPON RECEIPT OPEN THE SHIPPING CONTAINER AND INSPECT THE PLANTS. The tree seedlings are wrapped in moist paper and sealed in a plastic bag. If the paper wrapping appears dry, open the bag and thoroughly soak the paper with aged room temperature water. These are bare-root plants, which means that they have had all the soil removed from their roots. If the roots dry, the seedlings will be badly

damaged and may die. If the wrapping paper is moist, leave the tree seedlings in their unopened shipping bag until shortly before they are to be used.

FAQs

Q. How long can I keep the tree seedlings before using them?

A. If possible, use them within 2 to 3 days of receipt. See your Teacher's Guide for instructions for preparing the terraria.

Q. Can the seedlings be stored in a refrigerator?

A. This isn't recommended because of the danger of freezing. If you do refrigerate the seedlings, place them in the vegetable bin or another area of the refrigerator with little risk of near-freezing temperatures. This should be an emergency procedure only. It is much better to prepare the terraria and plant the seedlings as soon as possible.

Q. Why are the tree seedlings shipped bare-rooted?

A. Bare-rooted tree seedlings can be shipped in a dormant or resting state. They often survive shipment much better than seedlings in pots do.

Q. Are there any special precautions to take with bare-rooted plants?

A. Keep the roots moist at all times. Don't plant the seedling in dry soil. Premoisten the woodland terrarium soil as directed in your Teacher's Guide. When you plant the seedling, be certain that its roots make good contact with the soil. Don't leave any air pockets around the roots or they will dry.

VERTEBRATES

Dwarf African frog

(Hymenochirus boulengeri)

Uses: Study of the characteristics of animals, characteristics of vertebrates, consumers in aquatic food webs and chains, aquatic habitat, behavior

Level: Moderate

Unit(s): STC®: *Animal Studies*, Grade 4

Special requirements: Aquaria with aged and conditioned water



Frogs are amphibians, which is Greek for “double life.” They undergo metamorphosis, changing from an egg to a tadpole to a young frog. The dwarf African frog, also known as the dwarf aquarium frog, is strictly aquatic and found primarily in pools or sluggish water. These small frogs, which grow to about 1 1/2 inches in length, are gray or brown with a darker marbling pattern on their backs and light-colored abdomens. They have webbed feet, necessary for an aquatic existence, with claws on the 3 inner toes of each hind foot. Dwarf aquarium frogs have

small, lidless eyes, and although it is difficult to distinguish the sexes, the male is usually smaller and develops a yellowish swelling just behind its forelimbs during breeding.

Aquaria: Follow the directions in your Teacher’s Guide for setting up the aquaria. Treat all water for holding tanks and aquaria with the water conditioner included in your kit. Give some thought to where you will keep the aquaria. It is best to select an area where the temperature is fairly constant, between 18° to 25° C (64° to 77° F). An aquarium needs diffuse light and should not be placed in direct sunlight or unusually bright artificial light. Remember, frogs are cold-blooded animals. An aquarium in direct sunlight may have a temperature as much as 5° C above the temperature of the room. At night such an aquarium cools rapidly, stressing the frogs and leaving them vulnerable to disease.

Acclimating and holding dwarf African frogs: IMPORTANT! IMMEDIATELY ACCLIMATE FROGS TO HOLDING PAILS OR HABITATS—DO NOT LEAVE THEM IN THE SHIPPING BAGS! Refer to Appendix A in the *Animal Studies* Teacher’s Guide. Prepare 2 holding pails according to the instructions in Appendix A. Acclimate frogs to temperature and pH by removing one cup of shipping water and replacing it with one cup conditioned, room temperature tap water. Repeat this step after 15 minutes; allow the shipping container to sit undisturbed for 15 additional minutes. Gently transfer frogs from shipping water to 2 holding pails using an aquarium net. Hold your hand over the net to prevent the frogs from jumping out. *Elodea*, a water plant, is shipped with the frogs. Put half the plants into each holding pail. Punch air holes in the holding pail lids and cover the pails. Feed the frogs according to instructions in Appendix A. DO NOT OVERFEED! Keep them at room temperature and in indirect light. Do not further disturb the frogs until you are ready to use them for lessons.

FAQs

Q. What should I do with the water in the shipping bag?

A. Discard all the shipping water. It contains waste products given off by the frogs during shipment. If you add the shipping water to an aquarium, you are also adding these waste products. Flush the shipping water down a sink with tap water.

Q. The shipping bags are cold and the frogs aren’t moving. Are they dead?

A. The frogs are cold-blooded and become less active when they are cold. Allow time for them to warm to room temperature. Never apply heat or add warm water to hasten this process. A rapid change of temperature can do much more damage to the frogs than chilling them can.

Q. The frogs are probably hungry, so I should feed them, right?

A. The frogs need time to recover and become familiar with their new environment. Wait a day before feeding them.

Q. What should I feed the frogs?

A. Use the brine shrimp flakes as recommended in your Teacher’s Guide. We do not recommend other foods.

Q. How much food should I give the frogs?

A. Less than you think. People have a tendency to overfeed them. Follow the feeding schedule in your Teacher's Guide. Feed frogs only as much as they can eat in a few minutes. Any food that the frogs do not eat may become food for bacteria, and too many bacteria in the aquarium may use up the oxygen in the water. This stresses the frogs and other animals in the aquarium.

Q. How do I know if my frogs are healthy?

A. The frogs should be observed daily and any change in behavior or appearance noted. A white spot on the skin that gradually grows larger may indicate a fungal infection. A diseased frog should be removed before it transmits the disease to other frogs in the aquarium.

Q. How can I keep my frogs healthy?

A. The period following the introduction of frogs into an aquarium is critical. Acclimate your frogs properly, and they will be off to a good start. Maintain the aquarium to keep them healthy. Once each month, remove about 1/4 of the aquarium water and replace it with fresh water that has been aged and treated with water conditioner. This dilutes any toxic buildup in the aquarium and keeps the pH and ionic balance of the water within a normal range. Never replace more than 1/4 of the water at any one time. Avoid sudden changes to the aquarium. Stressed frogs often become sick frogs, so keep their environment stable and fresh.

Q. All the frogs died soon after we put them in the aquaria. What went wrong?

A. Did you properly acclimate the frogs? Did you use the water conditioner? Some city water systems now use chlorinators that are not removed by aging the tap water. In those cities, the water conditioner must be used to dechlorinate the water. If you acclimated the frogs properly and used the water conditioner, your tap water may contain metal ions that are toxic to frogs. This is especially likely if the water pipes at your school are less than 3 years old or if there have been recent, major repairs to the plumbing. You may have to use water from another source or bottled water. Finally, the frogs may have been overstressed during shipping. Request a replacement from Carolina Biological Supply Company.

Fish

(*Gambusia affinis*, *Carassius auratus*)

Uses: Study of the characteristics of animals, characteristics of vertebrates, consumers in aquatic food webs and chains

Level: Moderate

Unit(s): STC®: *Organisms*, Grade 1; *Ecosystems*, Grade 5

Special requirements: Aquaria with aged and conditioned water



Coming in a variety of shapes, sizes, and colors, fish are the most popular of aquarium animals. They are good examples of vertebrate body structure. *Gambusia* (mosquito fish) and guppies are livebearers and reproduce readily in a classroom setting.

Aquaria: Follow the directions in your Teacher’s Guide for setting up the aquaria. Treat all water for holding tanks and aquaria with the water conditioner included in your kit. Give some thought to where you will keep the aquaria. It is best to select an area where the temperature is fairly constant, between 18° to 25° C (64° to 77° F). An aquarium needs diffuse light and should not be placed in direct sunlight or unusually bright artificial light. Remember that fish and other aquarium animals are cold-blooded. An aquarium in direct sunlight may have a temperature as much as 5° C above the temperature of the room. At night such an aquarium cools rapidly, stressing the fish and leaving them vulnerable to disease.

Acclimating fish: IMPORTANT! IMMEDIATELY ACCLIMATE FISH TO HOLDING PAILS OR HABITATS—DO NOT LEAVE THEM IN THE SHIPPING BAGS! Allow 50 to 60 minutes to acclimate your fish. Extra time spent now heads off serious problems, so resist the temptation to rush. Read the relevant sections of your Teacher’s Guide. Your aquaria should be ready with conditioned water that is at room temperature. Avoid the 3 deadly stresses: temperature shock, pH stress, and toxin buildup.

Immediately upon arrival of the fish, open the top of the bag and let in some fresh air. Then either float the bag in the holding tank or simply allow it to come to room temperature (propped in a container to prevent spilling). After 20 to 30 minutes, remove and discard about 1/4 of the water from the bag and replace it with water from the holding tank or with other prepared water (aged or dechlorinated) from the same supply as that in the holding tank. Wait 15 minutes and repeat the above step. After another 15 minutes, carefully remove the fish with a net and place them in the holding tank. The water in the shipping bag has accumulated wastes and should be discarded. For more detailed instructions on fish care and water preparation, see your Teacher’s Guide.

FAQs

Q. What should I do with the water in the shipping bag?

A. Discard all the shipping water. It contains waste products given off by the fish during shipment. If you add the shipping water to an aquarium, you are adding these waste products. Flush the shipping water down a sink with tap water.

Q. The fish are probably hungry, so I should feed them, right?

A. The fish need time to recover and become familiar with their new environment. Wait a day before feeding them.

Q. How much food should I give the fish?

A. Less than you think. People have a tendency to overfeed fish. Feed the fish only as much as they can eat in a few minutes. Don’t feed them more than once a day. Any food that the fish do not eat may become food for bacteria, and too many bacteria in the aquarium may use up the oxygen in the water, stressing the fish and other animals in the aquarium.

Q. How do I know if my fish are healthy?

A. Fish should be observed daily and any change in behavior or appearance noted. A healthy fish usually keeps its fins erect and spread and is active. A fish with drooping fins or fins tightly

pressed against its body may be diseased. A fish that is always hidden may also be diseased. Watch for any fuzzy patches growing on the fish or long streamers of slime that trail from its fins or body. A diseased fish should be removed before it transmits the disease to other fish in the aquarium.

Q. How can I keep my fish healthy?

A. The period following the introduction of fish into an aquarium is critical. Acclimate your fish properly, and they will be off to a good start. Maintain the aquarium to keep them healthy. Once each month, remove about 1/4 of the aquarium water and replace it with fresh water that has been aged and treated with water conditioner. This dilutes any toxic buildup in the aquarium and keeps the pH and ionic balance of the water within a normal range. Never replace more than 1/4 of the water at any one time. Avoid sudden changes to the aquarium. Stressed fish often become sick fish, so keep their environment stable and fresh.

Q. All the fish died soon after we put them in the aquaria. What went wrong?

A. Did you properly acclimate the fish? Did you use the water conditioner? Some city water systems now use chlorinators that are not removed by aging the tap water. In those cities, the water conditioner must be used to dechlorinate the water. If you acclimated the fish properly and used the water conditioner, your tap water may contain metal ions that are toxic to fish. This is especially likely if the water pipes at your school are less than 3 years old or if there have been recent major repairs to the plumbing. You may have to use water from another source or bottled water. Finally, the fish may have been overstressed during shipping. Request a replacement.

INTERNET RESOURCES

There are thousands of good Internet sites about plants and animals. Here are a few to get you started.

Carolina Biological Supply Company

(www.carolina.com)

Carolina offers online care guides for a number of living organisms. To view these, click on the “Care guides for plants and animals” icon along the left margin of the Carolina homepage. This section also features some animal care video clips (which require RealPlayer™ or QuickTime™ software). See also back issues of the *STC®Update* newsletter

(www.carolina.com/STC/publications.asp), the STC living materials section

(www.carolina.com/STC/live_materials.asp), and revised instructions for the STC® *Animal Studies* unit, which now uses millipedes instead of land snails

(www.carolina.com/STC/animal_studies_change.asp). Look around a bit—new information is constantly being added.

National Science Resources Center

www.nsrconline.org

The NSRC, developer of the STC® and STC/MS™ programs, has created a student/teacher Web site supplement for the *Organisms—From Macro to Micro* module (www.stcms.si.edu/second_four.htm). Visit this site for resources and information on module topics.

Aquatic organisms

www.livefoodculture.com

Lots of folksy articles about culturing aquatic invertebrates and plants. Good, practical methods that could be adapted to the classroom.

www.wetwebmedia.com

A great source of articles and FAQs on aquaria, fish, other aquarium animals, and plants. Includes marine, freshwater, and brackish water aquaria and organisms.

Butterflies and moths

csce.unl.edu/~scotch/samantha

A hobbyist's page with lots of good, practical information on various species of moths and butterflies and how to raise and care for them.

www.public.iastate.edu/~mariposa/homepage.html

Everything you ever wanted to know about painted lady and red admiral butterflies.

www.npwrc.usgs.gov/resource/distr/lepid/bflyusa/bflyusa.htm

What butterfly is that? Just click on the map for a list of butterflies found in your state. Includes photos of most species listed, life histories, and more.

Invertebrates

www.uky.edu/Agriculture/Entomology

Enter the name of an insect for a list of articles. Good source of information, some technical, some general.

whatcom.wsu.edu/ag/compost/Redwormsedit.htm

Describes how to use redworms to compost garbage.

www.geocities.com/millipedes_uk

A hobbyist's page with everything you could possibly want to know about millipedes.

insected.arizona.edu/uli.htm

Great source of information on the care of various arthropods, including insects. Includes a good selection of elementary science lesson plans that use arthropods. Highly recommended.

ADVISORY ON RELEASING OR DISPOSING OF ORGANISMS

The National Science Resources Center advises against the release of any organisms used in the STC® or STC/MS™ program. In some documented cases, environmental problems have resulted from the introduction of nonindigenous organisms into nature. It is also illegal in many states to release organisms, even indigenous species, without a permit. (The cultured organisms

may carry genes that could affect the wild population.) The intention of these laws is protection of native wildlife and the environment.

The organisms used in the STC® and STC/MST™ units were thoroughly researched before they were selected. Other than the cabbage white caterpillars, the organisms are unlikely to harm local ecosystems. Nevertheless, allowing their release into nature might encourage your students to release other organisms that could cause harm to native wildlife and local ecosystems. If you have any questions about releasing organisms in your area, contact your state or local environmental conservation agency.

After you complete the unit, there are several things you can do with the organisms (except for the waxworm and cabbage white butterfly):

- Continue to maintain them in your classroom.
- Donate them to a pet shop, zoo, or nature center.
- Donate them to another classroom or to another school's science department.
- With parental permission, let your students take them home for use in personal aquariums, etc. As a last resort, biologists suggest that you place the organisms in a sealed container, freeze them overnight, and dispose of them according to your school district's policy on organic waste material disposal.

If you give the organisms to students or to other groups, please make them aware of the advisory on releasing organisms.

For the waxworm (greater wax moth) and cabbage white butterfly:

Waxworm: In nature, waxworms are detrimental to honeybees, which are important pollinators. The culture is easily maintained by following the instructions provided, so you could pass the culture along to another teacher. If this is not possible, destroy the waxworms by thoroughly freezing the culture.

Cabbage white butterfly: Because disposal of cabbage white caterpillars is regulated by the U.S. Department of Agriculture (see the special note on pages 81–82 in your *Organisms—From Macro to Micro Teacher's Guide*), disposal of the organisms must be controlled. Your 2 disposal options are:

- Wait until the butterflies die naturally. (The adult life span of the butterfly is short, about 2 weeks.) Consider saving the dead adult butterflies in a plastic cup with holes punched in the lid. They are ideal specimens to examine through the microscope.
- Humanely euthanize the eggs, larvae, and adult butterflies by placing them a sealed container in a freezer for 48 hours.

LIVING MATERIALS INFORMATION REQUEST FORM

If you have a question or need more information about any living material purchased from Carolina Biological Supply Company, use this form to fax or email your query to us. *Please do not use this form to request replacements of living materials or to place orders.*

FAX TO: **800.222.7112** or 336.538.6373 **OR** EMAIL TO: steve.binkley@carolina.com
ATTENTION: **Steve Binkley, Living Materials Department**
CAROLINA BIOLOGICAL SUPPLY COMPANY

FROM: _____ DATE: _____

SCHOOL OR DISTRICT: _____

YOUR FAX NUMBER, EMAIL ADDRESS, OR OTHER METHOD OF CONTACT: _____

NAME OF ORGANISM: _____

STC® OR STC/MS™ UNIT(S): _____

Write your question below, including all relevant information such as how long you have had the living material, conditions under which the living material is being maintained, etc.