INQUIRY & INVESTIGATION

An Inquiry-Based Field & Laboratory Investigation of Leaf Decay: A Critical Aquatic Ecosystem Function

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Abstract

Effective investigations incorporate all four features of constructivist teaching. This high school or college-level field investigation guides teachers (and students) through the stages of inquiry. The focal concept is ecosystem function, specifically leaf decay rates in aquatic environments. Teachers elicit their students' prior knowledge and use it to generate discussion on variables that influence decay rates. Students engage in designing and conducting experiments. The learning cycle is continued when students apply their new knowledge and receive feedback, and completed when students return to their initial conceptions of leaf decay and reflect on the knowledge they gained through scientific experimentation.

Key Words: *Constructivist; inquiry; ecosystem function; leaf decay; aquatic ecosystem.*

○ Essential Features of Constructivist Teaching

Effective inquiry-based investigations incorporate the four essential features of constructivist teaching methods: (1) eliciting prior knowledge, (2) creating awareness of differences between prior knowledge and new knowledge, (3) application of new knowledge with feedback, and (4) reflection on learning (Baviskar et al., 2009). The following investigation models this method, which has been demonstrated to foster critical thinking and reasoning, scientific literacy and understanding of the process of science, and the learning of science content (Shymansky et al., 1983; Lord, 1994, 1997, 1999; Bransford et al., 2000; Banet & Ayuso, 2003; Burrowes, 2003). This investigation into a critical ecosystem function addresses the following science education standards: "interdependence of organisms and biological change," "matter, energy, and organization of living systems," and "concepts of scientific inquiry" (National Research Council, 2000).

○ Overview of Ecosystem Functioning: Leaf Litter Decay

Ecosystems have both structure (i.e., number of species) and function (i.e., rate of energy flow). There are several tactile and visual ways in which biology students can learn about the structural aspects of ecosystems, such as collecting plants or insects to describe community composition (Barratt, 2004; Farone & Farone, 2005; Tomasek et al., 2005; Ruesink et al., 2006). There are fewer ways for students to learn about ecosystem functions. Leaf litter decay in aquatic ecosystems is a function that can be measured easily. Decay of leaf litter plays a pivotal role in stream food webs (Petersen & Cummins, 1974; Vannote et al., 1980; Allan, 1995) and represents the composite effects of biological, physical, and chemical activity. Students can design experiments and evaluate evidence from a variety of factors that influence decay rates. The results are highly visible and can be seen in a matter of a few weeks. This activity allows students to measure a critical ecosystem function in a real ecosystem, while understanding both structural and functional aspects of the food web in stream ecosystems (Figure 1).

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Leaves are a primary source of energy for streams and enter the water from the surrounding riparian area. Soluble nutrients in leaves leach into the water, and leaves are quickly colonized by microorganisms, especially fungi, in a process called conditioning (Figure 1). Several invertebrates, such as stonefly nymphs, caddisfly, and Diptera (fly) larvae, are dependent on these conditioned leaves for their food. These invertebrates are referred to as "shredders" because they fragment the leaves and facilitate the decomposition of this coarse particulate organic matter (CPOM) by breaking it up into smaller pieces, or fine particulate organic matter (FPOM). Leaves are also broken down by physical abrasion. The FPOM is the food resource for collector invertebrates, such as larval black flies. Both shredder and collector invertebrates are a food resource for higher-trophic-level organisms, such as predatory stoneflies and fish.

The process by which leaves decay in streams is amenable to experimentation by students. Leaves that fall into streams often cluster together in "packs" behind rocks and woody debris. Leaf packs can be constructed for experiments by collecting leaves from the surrounding riparian area. Students can calculate the rate of leaf decay by placing a known amount of leaf material in plastic mesh bags, placing the experimental leaf packs in a stream channel, and measuring how much leaf material remains after a set amount of time.

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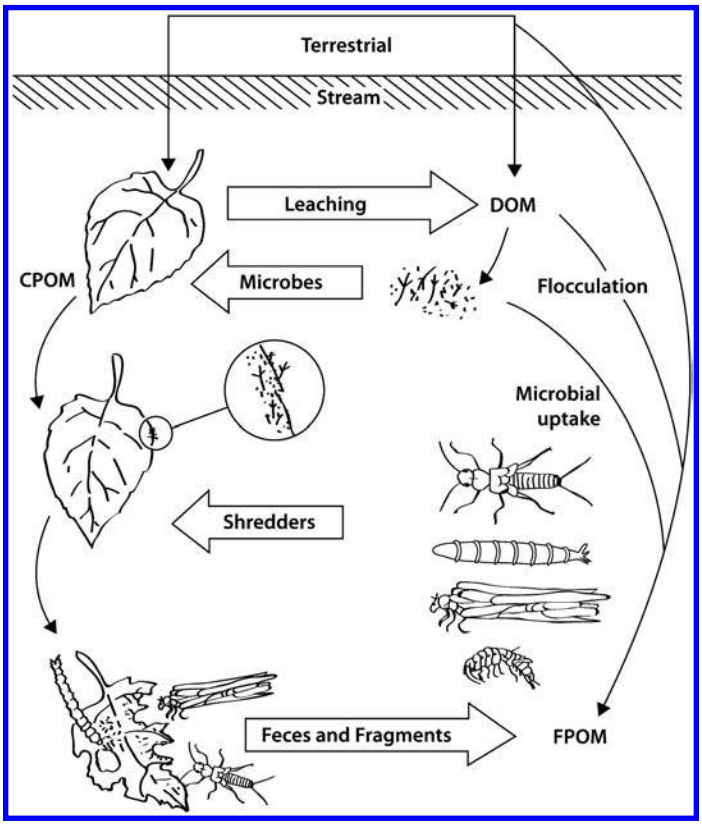

Figure 1. The sequence of leaf litter decay in streams. (Redrawn from Cummins & Klug, 1979.)

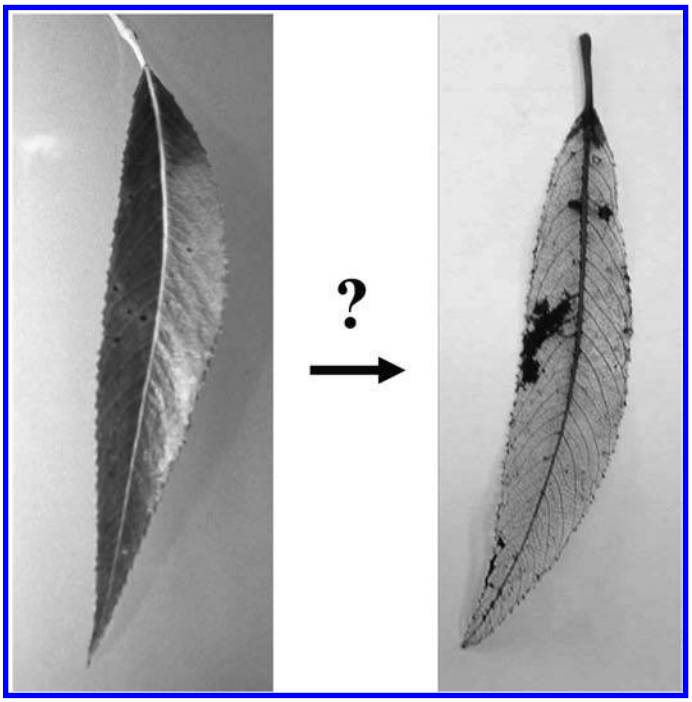

Figure 2. Asking students how an intact willow leaf turned into a decayed willow leaf can help elicit prior knowledge and design appropriate experiments to build upon what they already know.

Students measure an ecosystem function (the rate of leaf decay) and may simultaneously investigate ecosystem structure (species richness). Others have had success in teaching about ecosystem functions with an instructor-directed approach whereby students observe leaf decay in a laboratory (Sparkes et al., 2008). Our approach is student-directed inquiry in which the teacher acts as a guide and students measure the function in a real ecosystem.

○ Materials & Preparation

Equipment includes a drying area or drying oven, balances, spray bottles, builder's bricks, surveyor's tape or bright yarn, temperature data loggers or maximum/minimum recording thermometers, large buckets, white pans, sieves, forceps, and enough mesh bags with 30-cm-long cable ties for three per removal date per experiment or group. A muffle furnace is helpful for more precise measurements of leaf litter decay rates. Invertebrate analysis requires hand lenses, dissecting scopes, Petri dishes, and identification keys.

As part of the investigation, students can help identify stream locations that are accessible to the class and with sufficient flow to ensure that leaf packs will be submerged throughout the study. Depending on class size, objectives, and safety considerations, the students will need to return to the site several times for collection of the leaf packs.

○ Eliciting Prior Knowledge

To discover the students' present level of understanding about the process of leaf litter decay in streams, show them an intact leaf and a partially decayed leaf of the same type that was pulled from a stream channel (Figure 2). Ask the students to diagram what they think happened to make the intact leaf look like the decayed leaf. Encourage them to be as complete as possible in their descriptions. Collect their written responses as a record of their level of knowledge at the beginning of the lesson. Their responses will give you an idea of the level needed to challenge them and how to best address their misconceptions and add to their existing knowledge. Here, it is critical to recognize that not everyone has to do the same experiment. Guide your students to design experiments that challenge the gaps in their knowledge.

○ Getting Students to Ask Questions

During this part of the activity, guide the students to ask relevant questions about the process of leaf decay. Facilitate a group discussion that prompts them to identify factors that would affect how fast an intact leaf will decay. Initiate the discussion with questions, such as: Why does food go bad faster when it is left out rather than kept inside the refrigerator? (Temperature, microbes.) What might cause physical breakdown of leaves in streams? (Turbulence, abrasion.) A list of factors can be constructed that includes characteristics of the stream (e.g., temperature, dissolved oxygen, pH, nutrients, physical abrasion, stream biota) and characteristics of the leaf (e.g., cuticle thickness, lignin, tannin, and nutrient content).

As the students begin to see that the process of leaf decay in streams represents a combination of physical, chemical, and biological factors, have them think about how this process might be measured. Ask them to consider how they would compare decay rates of the same types of leaves in different streams. Typically, students quickly recognize that temperature plays a large role in the breakdown of organic matter. Since temperature is so critical, decay rates are normally standardized by adjusting for temperature and are reported as mass lost per degree-day. Degree-day is the sum

Table 1. Categories of leaf decay rates with examples of each classification. Categories are based on percent dry weight lost per degree-day: Slow <0.10, Medium >0.10 and <0.15, Fast >0.15. (Modified from Cummins et al., 1989.)

In-Stream Processing Category		
Fast	**Medium**	**Slow**
Alder	**Maple**	**Oak**
Dogwood	**Hickory**	**Conifers**
Basswood	**Willow**	**Aspen**

Table 2. Independent and dependent variables that can be used to design leaf-pack experiments. (F) = Functional measure. (S) = Structural measure.

Variables	
Independent	**Dependent**
Leaf type	Decay rates (F)
Leaf condition (from tree, ground, or stream)	Invertebrates (S)
Location in stream (pool, riffle, run)	Abundance/Diversity
Time left in stream	Microbes (S)
Presence/absence of invertebrates	Abundance/Diversity

of the mean daily water temperature for each day that the leaves were in the stream. The concept of degree-day will be new to most students and can be introduced early in the lesson and returned to later.

Students also should recognize that differences in leaf chemistry and structure result in a wide variation in decay rates among leaves of different species. It is well established that different types of leaves decay at different rates. One reason is that the concentration of tannins, which prevent decay, varies among species (for a laboratory investigation of tannins in leaves see Traw & Grift, 2010). Several common tree species have been placed in three broad categories; fast, medium, and slow decomposers (Table1). The students can collect leaves from several different tree species and make predictions about their decay rates in relation to other species. Known decay-rate values also can help you guide the students to design experiments that can be completed in the time available. If you only have 2 weeks to conduct experiments, you can encourage the students to choose leaves from the "fast" category.

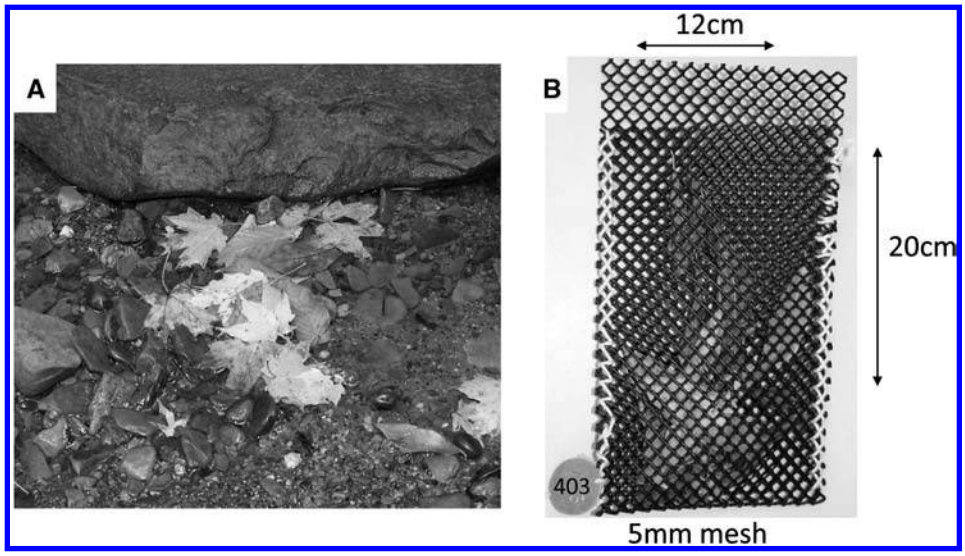

Figure 3. (A) Natural and (B) experimental leaf packs.

○ Investigate

During this phase of the inquiry cycle, the students design their own field experiments. Have the class organize the list of factors into independent variables that can be manipulated and dependent variables that can be measured (Table2). In order to design an experiment that can be successfully completed, the students should be exposed to the materials that are available and given the time constraints. Leaves collected directly from a tree or shrub in autumn just before they senesce are best. Those collected from the ground are preconditioned to some extent and may decay at a faster rate. Ideally, each experiment should have two or three removal dates in order to develop a decay curve. The removal dates can be predetermined using published decay rates as a guide. In general, all leaf packs are removed within 1 month after placement in the stream.

Introduce the students to the concept of experimental leaf packs as analogues for natural leaf packs (Figure 3). Mesh bags with labels can be purchased from LaMotte (item number: 5882-LPB, 30 bags for $20.50), or the students can use mesh bags saved from store-bought onions or other produce. These bags are not as durable and usually cannot be reused.

Have the students submit their research question, hypothesis, and study design in writing and provide feedback before they begin the experiments. There may be questions that are of particular interest locally (e.g., Do decay rates in urban and agricultural streams differ? Do leaves from exotic riparian plants break down faster or slower than those from native ones?). Experimental design will vary by individual student or group, but the general procedures for measuring leaf decay rates will be similar.

The following are general instructions for students.

Preparing Leaf Packs

1. Collect leaves from selected trees or shrubs, keeping them separated by species. Large brown paper bags work well for collection and storage.
2. Dry leaves to a constant dryness to standardize the water content of the leaves. This can be done at room temperature or in a drying oven at 50°C (recommended for humid locations). Drying will take a few days at room temperature or 24 hours in a drying oven.

3. Prepare the mesh bags and assign each one an individual number on a waterproof label attached to the bag.
4. Weigh leaves using a balance accurate to two decimal places. You will need 5–10 g of leaf material for each pack. Keep the amount relatively constant among packs within a given experiment. Record the weights of the leaves put into each leaf pack and the corresponding bag identification number.
5. Moisten the weighed leaves to reduce breaking and loss of leaf material. Place the leaves in a plastic bag, use a spray bottle to moisten them, and allow the leaves to remain inside the plastic bag for several hours or overnight.
6. Put the moistened leaves in the corresponding numbered mesh bag. Fold over the top and secure each leaf pack to a brick. One large cable tie can work to both close the opening in the pack and secure it onto the brick.

Placing Leaf Packs in the Stream

Measure flow velocity before placing leaf packs in the stream, using a flow meter if available. Alternatively, flow velocity can be estimated by measuring the length of time that a floatableobject (such as a piece of cork or an orange) takes to travel a known distance downstream.

1. Place the leaf packs in the stream with the leaves facing into the current so that as much surface area of the bag is facing the current as possible. Make sure that all leaf packs are submerged and securely anchored to bricks.
2. Secure a temperature logger or maximum/minimum recording thermometer near (or on) the leaf packs.
3. Draw a site map in a field notebook that shows the location of each leaf pack in the stream using their individual identifiers (tag number). Leaf packs may become covered with sediment and algae or the water level may rise, making them hard to locate weeks later. It is helpful to mark the vicinity of the leaf packs by tying pieces of surveyor's tape or brightly colored yarn to the surrounding vegetation.
4. Record date, time, and other relevant field measurements.
5. Keep the leaf packs in the stream for 1–4 weeks, depending on removal dates for your experiment. If possible, check the packs more frequently to see that they remain submerged. If using maximum/minimum recording thermometers, they can be read and reset each time you return to the stream.

Field Processing

Disassembling and sorting the leaf packs in the field is preferable to processing them in the laboratory because of the space constraints imposed indoors and the option to return live invertebrates to the stream. If processing in the field is not possible, all leaf bags can be placed in plastic freezer bags and returned to the laboratory in a cooler on ice. The packs can be frozen until sufficient time is available for processing. The methods for processing are the same, whether performed in the field or laboratory.

1. Process one leaf pack at a time. Pick up the brick and leaf pack together and quickly place them in a 5-gallon plastic bucket partially filled with stream water. Cut the cable tie and take the leaf pack off of the brick. Set the brick aside.
2. Open the mesh bag and shake the contents into the bucket.
3. Rinse the contents of one leaf pack into a bucket. The material in each bag should be rinsed of sediment, invertebrates, and extraneous detritus.
4. Separate the leaves from everything else in the bucket. Pick up a few leaves at a time and agitate them to dislodge macroinvertebrates. Transfer those leaves to a large white enamel pan with some stream water.
5. If collecting macroinvertebrate data, pour the contents of the bucket through a strainer and preserve the invertebrates for later identification. Otherwise, the invertebrates can be returned to the stream.
6. Repeat for each leaf pack. Make sure to label and keep the materials from each leaf pack separate.

Laboratory Processing

1. Spread the rinsed leaves out for drying. Keeping each leaf-pack sample separate, either place them in a drying oven set at 50°C, spread them out onto newspaper, or place in stackable mesh sieves and allow to dry overnight.
2. After drying to a constant weight, record the weight of the dried leaf material along with the leaf-pack identification number. Be sure to subtract the weight of the weighing dish for each sample.
3. If collected, invertebrates can be classified to order or family level using simple dichotomous keys and other identification aids. Several area-specific field guides exist, and many websites offer excellent photographs to help identify invertebrates. Record the leaf pack number and the invertebrate data on a separate data sheet.

○ Linking Prior Knowledge to New Knowledge

Depending on the design of specific experiments, the students will perform several calculations and analyses in order to answer their specific questions. As time permits, results can be compiled for comparisons between different leaf types or locations. In general, analyses are based on dry weights of leaves before and after they were placed in the stream. The students can directly analyze the basic calculations of percentage of leaf material lost per degree-day. More advanced students can calculate the decay rate coefficient and test for significant differences using parametric statistics (see Minshall & Rugenski, 2007). All the students should be guided to think critically and logically about the evidence provided through their analyses and create their own explanations.

Basic Calculation of Mass Lost

Mass lost = initial dry mass – final dry mass
Percentage remaining = (final dry mass / initial dry mass) $*$ 100
Percentage lost = 100 – percentage remaining

Degree-Day

Calculate the mean daily water temperature for each day that the leaf pack was in the stream and sum means to get the degree-day for that leaf pack. To correct decay rates for temperature, divide the mass lost by degree-day. This will allow for comparison of sites, species, and seasons.

○ Discuss & Present (Application of New Knowledge with Feedback)

Have the students communicate their results. Excellent descriptions of alternatives to the familiar lab report can be found in the literature, including the use of posters (Billington, 1997), scientific symposia (Marcum-Dietrich, 2010), and scientific journaling and poster sessions (Shane, 2008). The important aspect is that the students model how scientists communicate, by sharing what they have learned with

their peers and expecting to question one another. This way, they will actively engage in scientific inquiry and make use of what they and others have learned.

○ Reflect & Revise (Assessment of Learning)

Finally, ask the students the same question that you posed at the beginning of the lesson when you elicited their prior knowledge. Have them diagram the process of in-stream leaf decay again after their investigations. Once they have completed their diagrams, return the written responses that you saved from the first time you asked the same question. Have the students compare their present level of understanding to what they knew about the process at the beginning of the lesson. The students should be able to recognize more factors and linkages in the process and recognize that their understanding of the natural world can be changed through scientific investigation. At this point, facilitate a discussion that leads the students to ask more questions to build upon their knowledge, emphasizing that this is the process of science.

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