



*Procedures*

## GETTING STARTED

1. Introduce the LPN to your students.
2. Decide the purpose of your experiment.
  - a. Involve your students in deciding what will be studied. As part of Leaf Pack Network<sup>®</sup>, all groups should conduct experiments with control leaf packs. **Control leaf packs** contain leaves from the three most common trees around the stream and are placed in a riffle.

*Note: Control leaf packs are recommended and constitute the minimum involvement in the Leaf Pack Network<sup>®</sup>.*



- b. The next step may involve designing an **experimental leaf pack**. Experimental leaf packs allow groups to become creative and enhance their project by comparing the control leaf pack to an experimental variable. Your choice of variables might be:

- different types of leaves in the leaf pack (ie. different species, fresh leaves, non-native species)

**OR**

- location of the leaf pack (ie. pool vs. riffle, forest stream vs. meadow stream)

- c. Have students develop a hypothesis about each leaf pack.
3. Decide on the location for the Leaf Pack Experiment and complete the **Site Data Sheet**. To determine latitude and longitude of your project use a GPS unit or visit:
    - a. [www.maptech.com](http://www.maptech.com) and at the top of the page select on-line maps, then MapServer.
    - b. <http://terraserver.microsoft.com/>
  4. First-time users must register as Leaf Pack Network<sup>®</sup> participants by completing the **On-line Application Form** at [www.stroudcenter.org/lpn/getinvolved.htm](http://www.stroudcenter.org/lpn/getinvolved.htm). Once your registration has been approved, start a new project. Remember, start a new project for each set of leaf packs (ie. Experimental vs. control leaf packs).

### General Tips:

**Habitat Data Sheet:** *If students cannot make it streamside to fill out the data sheets, take digital photos or make a short video of the leaf pack stream site, then show them in class. Make sure to label photos upstream, right/left bank, and leaf pack site. Use a topo map of the leaf pack site to further enhance your students' ability to fill out the habitat sheet and determine land-use.*

**Lab Procedures:** *Instead of transporting stream water back to the classroom, leave 3 buckets of tap water in a cool (but not freezing) place for 3 days before you retrieve your leaf packs. Chlorine will evaporate and when finished, macroinvertebrates with a little water can be returned to the stream. Local monitoring groups, like Trout Unlimited can be contacted for assistance.*

## PREPARING LEAF PACKS FOR THE STREAM

### Materials:

(\* included in Leaf Pack Experiment kit)

mesh bags\*

scales\*

tree identification guide\*

waterproof marker\*

scissors

waterproof tags\*

string/nylon twine\*

paper bags

### Procedures for the control packs:

1. Identify the three most common trees or shrubs near your stream.
2. Collect and separate leaves.

#### Tips:

- Before collecting leaves, weigh 30 grams of leaves as a demonstration so that students will understand how many leaves are required.
  - You may pick leaves off the plant or gather fallen leaves from the ground. Lay them out to dry if they are not already in that state and temporarily store the leaves in an open paper grocery bag if necessary.
3. Prepare three mesh bags for the control leaf packs. Use the scale to weigh 30 grams of dry leaves (approximately 10 grams for each leaf type) for each leaf pack. *Optional:* Prepare three mesh bags for the experimental leaf packs.
  4. Complete the tag for each mesh bag using the waterproof marker. Place the waterproof tag in the leaf pack bag. Include the following information:
    - date
    - bag number
    - identify as control leaf pack or experimental leaf pack
    - location
    - school/group name
  5. Tie each bag closed.
  6. Loop a long length of nylon twine or string through the mesh of each bag so the leaf pack can be attached to a large rock in the stream.

## PLACING LEAF PACKS IN THE STREAM

### Materials:

thermometers\*  
prepared leaf packs  
Site Data Sheet or blank paper

string/nylon twine\*  
Field Data Sheet

pencil  
Habitat Data Sheet

### Procedures:

*Note: If you are Determining Stream Discharge, do so before placing leaf packs. Follow procedures found in Learning Activities section.*

1. Tie leaf packs directly to existing rocks in the stream (when possible, in a riffle habitat). If there are no rocks, use overhanging roots or use cinder blocks with reinforcing rods (rebar's). Use a sledgehammer to drive a piece of rebar through the hollow part of each cinder block to secure it to the streambed. *Note: Please remember to remove these when your project is completed.*
2. Position leaf packs (facing upstream) so that as much surface area of the bag is facing the current as possible. Make sure all leaf packs are submerged and securely tied. *Note: Leaf packs that flap with the current are not properly placed.*
3. Record appropriate information on the ***Field Data Sheet***.
4. Draw a Site Map on the back of the ***Site Data Sheet*** that shows the position of each leaf pack in the stream. Leaf packs may become covered with sediment and algae making them hard to locate weeks later. *Note: It may be useful to place markers along the bank of the stream. However, they may draw attention to your leaf pack and possibly be vandalized.*
5. Complete the ***Habitat Data Sheet***. Refer to glossary for definitions.
6. Keep the leaf packs in the stream for 3-4 weeks. If possible, check the packs periodically to see that they remain submerged.

## COLLECTING LEAF PACKS FROM THE STREAM

### Materials:

site map	Field Data Sheet	zipper-top bags*
thermometer*	cooler with ice	scissors
bucket		

### Procedures:

1. Complete the remaining information on the *Field Data Sheet*. (For example: time, air and water temperature, number of packs and date removed, precipitation/storm information.)

*Note: Refer to the Site Map to identify the location of each leaf pack. Be sure to collect leaf packs starting with the one furthest downstream then work your way upstream.*

2. Collect a few inches of stream water in the bottom of a zipper-top bag.
3. Have one student gently hold onto the submerged leaf pack while another student cuts the twine securing it to the rock or block.
4. Pick up the leaf pack quickly and gently. Place it into the zipper-top bag and seal. Some of the insects are very quick and will try to escape.
5. Collect additional stream water in a bucket so that it can be used during macroinvertebrate sorting and keep cool. Tap water, allowed to dechlorinate for 3 days, can be used as well.
6. Place each zipper-top bag into a cooler to bring the leaf packs indoors or process leaf packs streamside. Most aquatic insects can be refrigerated or kept in coolers with ice overnight. Some invertebrates are very sensitive to changes in temperature. Therefore, try to keep them consistently cool.

*Note: Suggestions on how to maintain the macroinvertebrates overnight:*

- Place in a cooler with stream water and leave outdoors if cool (<15°C, keeping out of direct sunlight) or put freezer packs in cooler for indoor storage.
- Keep trays on a plastic bag filled with ice in the classroom.
- Use an insulated bucket (bait bucket) and add an air-stone.
- Store bags in a refrigerator.

## LAB PROCEDURES

### Materials:

Team Data Sheet	strainer*	spoons*	trays*
Leaf Pack Summary Sheet	hand lenses*	sorting sheets*	brushes*
Project Summary Data Sheet	petri dishes*	bowls	scissors
macroinvertebrate flashcards*	identification keys	squirt bottle	3 buckets

### Processing leaf packs:

*Note: We recommend processing leaf packs streamside. Process one leaf pack at a time.*

1. Cut or untie the twine. Place leaf pack contents in a bucket three-quarters filled with stream water. Rinse remaining contents of zipper-top bag in the bucket.
2. Vigorously shake small handfuls of leaves in the bucket to dislodge macroinvertebrates. Dispose of those leaves in a second container or zip-lock bag with some stream water and bring to classroom for later examination.
3. Pour contents of the bucket through strainer into another bucket. Add strainer content to trays (steps 4-5) or place in zip-lock bag with some stream water and bring back to classroom.

### Preparation for sorting:

*Note: Divide one leaf pack among the students, keeping the other sorted leaf packs in a cooler or refrigerator. Organize class into groups. Consider steps 4-5 as classroom preparation.*

4. Fill each group's tray with 1/2 inch of stream water. Distribute contents of the strainer among each group's tray. *Note: It may be helpful to squirt water through the back of the strainer into trays to dislodge the macroinvertebrates. Check the second container for any missed macroinvertebrates.*
5. Repeat for each leaf pack. Make sure to keep leaf packs separate and properly labeled.

*Note: If the tally from the first leaf pack exceeds 100 macroinvertebrates, you may report that one leaf pack **if** you do not have time to process the other leaf packs. Ideally, all packs should be tallied with individual data sheets.*

## LAB PROCEDURES (cont.)

### Sorting:

*Note: Each group should have a copy of the **Team Data Sheet**.*

1. Place a petri dish with stream water on each circle of the sorting sheet found in the Leaf Pack Experiment Kit.
2. Using a brush or spoon, place each macroinvertebrate in the petri dish next to the drawing it resembles. Place all macroinvertebrates that look alike in the same petri dish. Use hand lenses to check for the special characteristics of each macroinvertebrate. Use additional identification keys where needed.
3. Count the number of macroinvertebrates in each petri dish. Each group should complete a **Team Data Sheet**. *Optional: EPT Richness Worksheet Learning Activity.*
4. Combine data from each **Team Data Sheet** to the **Leaf Pack Summary Sheet**.
5. When all three leaf packs have been counted, place total from the **Leaf Pack Summary Sheet** onto the **Project Summary Data Sheet**.
6. Calculate the sum of the three leaf packs and place the total in the Total column. These are the numbers that are to be entered onto the Leaf Pack Network<sup>®</sup>. *Note: The Leaf Pack Network<sup>®</sup> will average the numbers based upon the number of leaf packs. For added math involvement have students calculate the average number of macroinvertebrates found per leaf pack.*
7. Enter data on the Leaf Pack Network<sup>®</sup>. *Optional: For student conducted calculations use Macroinvertebrate Analysis Worksheet Learning Activity.*

### Clean up:

- *Wash and dry all lab equipment before storing.*
- *Return macroinvertebrates to the place they were collected as soon as possible.*
- *Any macroinvertebrates that die can be preserved for future use in a reference collection if placed in 70% ethyl alcohol.*

## ENTERING DATA ON THE LEAF PACK NETWORK®

### *Are you a New User?*

Go to [www.stroudcenter.org/lpn](http://www.stroudcenter.org/lpn) and click on the “Get Involved” link. Follow the prompt to apply online as a Leaf Pack Network® Participant. Shortly thereafter you will become approved, by the Leaf Pack Network® Administrator, to upload data.

### *Established Users:*

1. Go to: [www.stroudcenter.org/lpn](http://www.stroudcenter.org/lpn)
2. Select from the left hand menu bar:  
**Participant log in:**  
**Click here!**
3. Enter your user name and password. Submit.
4. Select from the left hand menu bar: **Start a New Project.**
5. Enter data for type of project, project name, watershed, sub-basin, stream name, month and year the leaf pack project was done. Submit.
6. Use the drop-down menu to select a project to edit. Submit.
7. Enter data from the Site Data, Field Data and Habitat Data Sheets as complete as possible. Submit.  
*Note: You must enter the date the leaf packs were placed **and** removed before data becomes viewable to the general public.*
8. Select from the left hand menu:  
**Enter data for:**  
**Stream Site**  
**Macro Data**  
**Journal Entry**  
**Add Journal Photos**
9. Enter data from Summary Leaf Pack Sheet and Journal Entry. Submit.  
Make sure pictures you upload are below 200kb in size. Upload Image.
10. Log-out.
11. Archive data sheets for future reference and study.

## JOURNAL ENTRY & JOURNAL PHOTOS

### To make a Journal Entry

1. Make sure to match up the Journal Entry with the correct project name.
2. Write your Journal Entry and click submit when finished. HTML is allowed.
3. When completed, pictures can be added to this journal.



Submit Journal

### TO ADD JOURNAL PHOTOS:

1. Simply click on the "Browse" button and find the picture on your hard drive.
2. Double click on it and it should appear on the first line below.
3. Please label your picture as well.
4. Click "Upload Image". Your image will now appear at the bottom of the page.
5. You can add up to 4 photos but you must add one at a time.

*Note: Large pictures may take a long time to upload or they may cause your connection to time out resulting in an error. Please try to keep individual photos under 200 KB. If you have any difficulties please email the Leaf Pack Network<sup>®</sup> Administrator at [leafpacknetwork@stroudcenter.org](mailto:leafpacknetwork@stroudcenter.org).*

Add Picture:

Label:

Upload Image