ISSUES : DATA SET  
  
**Investigating Leaf Litter Decomposition and Invertebrate Communities in Streams**

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*Survey flag marking sample site in stream. (Photo by T. Snukuts)*

**THE ECOLOGICAL QUESTION:**

How do abiotic and biotic factors impact invertebrate species diversity, invertebrate community structure and leaf litter decomposition in streams?

**ECOLOGICAL CONTENT:**

Species tolerance curves, diversity indices, litter decomposition rates, invasive species, and freshwater ecology

**WHAT STUDENTS DO:**

This dataset exercise has been developed from a classroom-based undergraduate research project on stream ecology that has been conducted in a second-year introductory ecology course. In this exercise, students first collect background information on what factors affect benthic macroinvertebrate communities or leaf litter decomposition in streams. Based on their acquired knowledge, students then develop literature-based hypotheses independently, or as a group. Students then examine the supplied dataset, plot figures and analyze data relating to their hypotheses. Students hand-in a scaffolded set of assignments that reflect different portions of a scientific paper. This format allows instructors to provide formative feedback from the students’ initial reading of the literature to their final analyses. Alternatively, instructors may select to have students plot figures of specific relationships to highlight certain ecological concepts that pertain to community or ecosystem ecology. This activity can be modified to suit a variety of levels.

**STUDENT-ACTIVE APPROACHES:**

Independent or group open-inquiry projects

**SKILLS:**

* *Hypothesis Formation*: develop literature-based hypotheses and predictions
* *Data Analysis*: select data from a series of Excel worksheets for analysis, conduct basic statistical analyses
* *Data Visualization*: create figures to visually represent data
* *Data Interpretation*: interpret results and evaluate hypotheses

**ASSESSABLE OUTCOMES:**

Annotated bibliography, proposed hypotheses and data analysis,written interpretation of figures and statistical analyses

**SOURCE**:

Data were obtained from a classroom-based undergraduate research project developed and taught by A.F. Janmaat.

**ACKNOWLEDGEMENTS:**

First and foremost, I would like to acknowledge all the students who have participated in the stream study, since its inception in 2008, and in particular those students who participated in the study in 2015. I would like to acknowledge Barbara Moon for including Streamkeepers training in the first years of the ecology course which provided the framework for the stream study. I would like to thank Sharon Gillies for her continued support of the project and for co-teaching the stream study for two years. I would like to thank Vicki Marlatt, Allan Ardnt, and Jenn Barrett, who each co-taught one semester of the study. I would like to thank Leslie Wood, Andrea Muelchen, Avril Alfred, and Alexa Kernel for technical support. I would like to thank Lucy Liu and the Faculty of Science for their support of the project. Funding for the stream study was provided by the Faculty of Science, UFV. I would like to thank the members of the QUBES Dig into Data Faculty Mentoring Network who provided feedback on the dataset and two anonymous reviewers.

**OVERVIEW OF THE ECOLOGICAL BACKGROUND**

Leaf litter is often the primary source of carbon within streams and the breakdown of leaf litter through decomposition is the foundation of most temperate stream energy webs. Decomposition in streams is primarily achieved by the combined action of physical factors, bacteria, fungi and invertebrate detritivores. Environmental factors such as temperature, pH, and nutrient levels may impact the rate of decomposition through their impact on these decomposing organisms. Therefore, leaf litter packs and their decomposition rates have been suggested as a way to monitor biological processes within local streams (Gessner and Chauvet, 2002). Changes in the decomposition rate affect the energy available within the stream system, and ultimately the community of organisms living there.

In addition to environmental factors, the quality of the leaves as a food source impacts the decomposition rate (Leroy and Marks, 2006). Leaf litter quality varies with the leaf litter species and will differ among native and invasive species. Accordingly, areas invaded by exotic species often differ in their decomposition rates from areas containing predominantly native species (Ehrenfeld 2010). It is, therefore, important to examine the decomposition rates of invasive species relative to the local native species to gain a better understanding of the impacts of the invasion on ecosystem processes.

The provided dataset is from a classroom-based undergraduate research project conducted by students at the University of the Fraser Valley (UFV) in British Columbia, Canada. In the study, students placed artificial leaf packs in streams near the Abbotsford campus of UFV. Subsets of the packs were collected at biweekly intervals, colonizing invertebrates were removed and identified, and the remaining leaf material was dried and weighed. In addition, the depth, temperature, pH and nutrient (nitrate and phosphate) concentrations of each of the stream sites were measured.

There is an extensive body of literature on leaf litter decomposition in streams and the associated macroinvertebrates. Access to this primary literature provides an opportunity for students to explore current research findings and develop diverse hypotheses that are supported by the literature and are testable with the provided dataset. The series of assignments outlined below, provides students with an open-ended inquiry of the impacts of biotic and abiotic factors on leaf litter decomposition and/or benthic macroinvertebrate communities. Alternatively, faculty may wish to direct students to look at specific relationships. Example comparisons that effectively illustrate ecological concepts, including species’ tolerances, community structure, and decomposition, are listed in the faculty notes.

**REFERENCES**

Ehrenfeld, J.G. 2011. Ecosystem consequences of biological invasions. Annual Review of Ecology, Evolution and Systematics. 41:59-80.

Gessner, M.O. and E. Chauvet. 2002. A case for using litter breakdown to

assess functional stream integrity. Ecological Applications 12:498-510.

LeRoy, C.J. and J.C. Marks. 2006. Litter quality, stream characteristics and litter diversity influence decomposition rates and macroinvertebrates. Freshwater Biology 51:605-617.

**DATA SETS**

* [Stream Study Data Student Version.xlsx](http://tiee.esa.org/vol/v13/issues/data_sets/janmaat/resources/Stream%20Study%20Data%20Student%20Version.xlsx)
* [Stream Study Data Faculty Version.xlsx](http://tiee.esa.org/vol/v13/issues/data_sets/janmaat/resources/Stream%20Study%20Data%20Faculty%20Version.xlsx)

The Stream Study Data Student Version is provided in a separate attachment and includes five Excel worksheets. Two worksheets include the benthic macroinvertebrate data separated by site, or by site and leaf litter species. A third worksheet includes the leaf litter decomposition data. A fourth worksheet includes site-specific environmental data and the final worksheet includes results of leaf litter tissue analyses. Notes are provided with each dataset that provide more details on how the data were collected.

The Stream Study Data Faculty Version includes a series of Excel worksheets that correspond to each of the comparisons that are discussed in the faculty notes. Each worksheet includes a datset specific to the comparison that was constructed using the student dataset and associated example figures.

Permission for this dataset to be posted and distributed on the TIEE website is provided by the author, Alida F. Janmaat. All data provided was collected by students enrolled in laboratory sections taught by A.F. Janmaat in 2015, and were collected according to procedures designed by A.F. Janmaat. Collection of the data was funded by the University of the Fraser Valley, Faculty of Science.

**STUDENT INSTRUCTIONS**

Leaves are often the primary source of carbon within streams and the breakdown of leaf litter through decomposition is the foundation of most temperate stream energy webs. Decomposition in streams is primarily achieved by the combined action of physical factors, bacteria, fungi and invertebrate detritivores. The rate of decomposition is affected by many factors, including temperature, nutrients, macroinvertebrate communities, and the nature of the leaf litter. Streams in urban and rural environments are subjected to a number of stressors that include altered flow regimes, high nutrient inputs, reduction of canopy cover, and the introduction of invasive species. Leaf-litter breakdown rates can be used as a tool to detect changes in these abiotic or biotic factors (Gessner and Chauvet, 2002).

Leaf litter decomposition in streams is most commonly measured using the litterbag technique. A known quantity of leaf litter is placed into a mesh bag, and the bag is then placed on the stream bottom. The mesh bags allow smaller aquatic invertebrates as well as microorganisms access to the leaves. Bags are harvested at periodic intervals, dried and reweighed to determine the amount of mass lost. Aquatic invertebrates are often removed from the leaf litter bags when they are harvested to provide information about the community of organisms inhabiting the stream. The composition of aquatic invertebrate communities has often been used to assess water quality, as many invertebrates are only able to survive within a limited range of environmental conditions. By assessing both the leaf litter decomposition and the invertebrate community, we gain a broader understanding of the impact of different factors on both community and ecosystem-level processes in streams.

Streams in the Fraser Valley region of British Columbia, Canada are facing many challenges due to the rapid urbanization of the area and intensive agriculture. Nitrate pollution is a significant factor in the water ways of the Fraser Valley (Mitchell et al., 2003), and urbanization is greatly impacting stream flow regimes (Hall and Schreier, 1996; Leith and Whitfield, 2000). Extreme weather events are predicted to increase with our changing climate, and in the Fraser Valley increasing summer droughts and severe rain events in the fall are predicted to increase in frequency and intensity (BC Agriculture and Food Climate Action Initiative, 2015). These changes will greatly impact streams in the Fraser Valley, the organisms that live in the streams, and ultimately the ecosystem processes that are the foundation of the stream communities. In order to capture the effects of the stressors impacting streams in the Fraser Valley, students from the University of the Fraser Valley have been studying leaf litter decomposition and associated macroinvertebrate communities in streams located near their campus. During the study, students place leaf litter bags at multiple stream sites, and measure the percentage of leaf litter remaining after 7, 14 and 28 days. The provided dataset includes the results of one semester of the ongoing study, during which litter bags were placed at 7 different stream sites (Table 1). The data provided were collected during a period of severe drought in 2015 when many of the streams were experiencing severely reduced flows.

Invasive plant species are also prevalent in riparian areas throughout the Fraser Valley. These species are now the source of leaf litter for many of the organisms that live in the streams. Leaf litter quality is one of the primary factors that impacts leaf litter decomposition and it varies considerably among native and invasive species. It is, therefore, not surprising that areas invaded by exotic species often differ in their litter decomposition rates from areas containing predominantly native species (Ehrenfeld 2010). In order to examine the impacts of invasive species in the riparian areas, UFV students have been constructing leaf litter bags from a variety of plant species typically found alongside streams. Two species are native to the riparian areas and include Thimbleberry (*Rubus parviflorus*) and Salmonberry (*Rubus spectabilis*). Two species are exotic to the region and include Himalayan Blackberry (*Rubus armeniacus*) and Japanese Knotweed (*Fallopia japonica*). These invasive species are among the most problematic exotic plant species in the Fraser Valley and can greatly impact riparian areas (Gaire et al. 2015; ISCBC, 2016). Twelve leaf litter bags from each species were placed at each stream site and four bags per species were attached to one of three bricks that were secured to the bottom of the each stream site.

Table 1: Stream Site Locations and Description of Riparian Habitat. Canopy cover and the presence of invasive species were measured at a minumum of 6 points at each studied location. Canopy cover was scored across a 5 m transect extending perpendicular to the stream at each sampling point by scanning overhead and rating cover according to a Braun-Blanquet scale.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Stream Site | GPS Coordinates | Canopy Cover [%] | Presence of Invasive Species  [% sampled points with species] | |
| Blackberry | Knotweed |
| 1 | 49.08313oN, 122.24987oW | 37 | 67 | 50 |
| 2 | 49.07901oN, 122.23772oW | 32 | 17 | 0 |
| 3 | 49.06228oN, 122.24709oW | 90 | 33 | 0 |
| 5 | 49.08103oN, 122.21488oW | 47 | 0 | 0 |
| 6 | 49.04621oN, 122.26053oW\* | 68 | 100 | 0 |
| 9 | 49.22192oN, 122.90074oW | 29 | 25 | 0 |
| 10 | 49.05620oN, 122.31160oW | 50 | 83 | 0 |

\* Site 6 was severely polluted due to the illegal dumping of milk into the creek by an upstream dairy farm which was discovered after the data was collected. Students noted a white substance in the creek on one occasion.

Outlined below are a series of assignments. In the first assignment, you will start by reviewing the literature relevant to the stream study dataset. You will then use the background information to develop a hypothesis that can be tested using the dataset in the second assignment. Once you have formulated your hypothesis and received feedback, you will outline how you will statistically test your idea in the third assignment. Finally in the last assignment, you will analyze your hypothesis and present your results.

1. **Background Literature Search**

To familiarize yourself with your research topic, you will need to briefly review the literature that pertains to the stream study. Find 2-3 representative papers from the peer-reviewed literature to introduce you to the topic. Your instructor may provide you with a general topic area (Table 1) that you may use as a guide during your search process to help you select appropriate papers. Read the papers and complete an annotated bibliography (**Assignment 1**). Your annotated bibliography should include a brief summary of each article, a sentence or two detailing the relevance of the article to the research topic, and another sentence that draws comparisons among the cited articles.

**Table 2:** Examples of general research topics and questions that can be addressed with the stream study dataset.

|  |  |  |
| --- | --- | --- |
| **Level of Ecology** | **General Topic** | **Sample Question** |
| Organismal | Species Tolerances | What is the effect of increases in nitrate concentration on the abundance of Ephemeroptera? |
| Community | Species Diversity and Abiotic Factors | Does macroinvertebrate biodiversity decrease as nutrient concentrations in the water increase? |
|  | Species Diversity and Biotic Factors | Does the colonization of leaf litter by macroinvertebrates differ among different leaf litter species? |
|  | Succession | Is there a specific order of macroinvertebrate colonization of leaf litter? (E.g. Shredders followed by collectors?) |
| Ecosystem | Decomposition and Abiotic Factors | Does leaf litter decompose faster at higher temperatures? |
|  | Decomposition and Biotic Factors | Does invasive plant leaf litter decompose more rapidly than native plant leaf litter? |
|  | Species Diversity and Decomposition | Does macroinvertebrate diversity increase leaf litter decomposition? |

1. **Formulating your Research Question and Hypothesis**

Now that you have read a number of scientific articles on your topic area, it is time to formulate your research question in **Assignment 2**. The question sets out what you hope to learn about the topic, and will guide and structure the choice of data to be analysed. What are the questions that you plan to answer using the data provided? Based on what is known in this field, what do you expect to see? From your research question, you will formulate your specific hypothesis/es. Your hypothesis/es will provide a tentative explanation for the relationships that you are expecting to observe in the provided dataset.

To aid you in the development of your hypothesis/es, a list of the variables provided in the stream study dataset is provided in Table 3 and a description of the stream study sites was provided in Table 1. Carefully consider the variables listed in Table 3. Which variables would you refer to as dependent variables? Which would you refer to as independent variables? Be sure to consider whether your hypothesis/es can be tested with the data provided. If your hypothesis/es cannot be tested with the available data, then you will not be able to complete assignments 3 and 4.

Your assignment should include a title, introduction with a brief literature review, and finally your hypothesis/es. The title should provide a very brief but specific summary of what you propose to examine. The Introduction should begin with the basics of your research topic and then further narrow the focus of those details to those that are most relevant to your hypothesis/es. In your introduction, present background information from the papers that you have read. You are laying the groundwork for your study with the summary of the papers that you present. The final part of the assignment is your research hypothesis/es.

**Table 3:** Variables measured during the Stream Study.

|  |  |
| --- | --- |
| Abiotic Factors | Variables were measured per stream site |
|  | pH |
|  | Water temperature [oC] |
|  | Water Depth [cm] |
|  | Nitrate concentration [NO3] [ppm] |
|  | Phosphates [PO4] [ppm] |
| Leaf Litter Quality | Variables were measured per litter species |
|  | Nitrogen [%] |
|  | Phosphorous [%] |
|  | Carbon [%] |
| Macroinvertebrates | Variables were pooled per stream site and collection date |
|  | Total Abundance |
|  | Identification (Family and Order) |
|  | Pollution tolerance (tolerant, moderate, sensitive) |
|  | Feeding type (collector/gatherer, shredder. Predator) |
|  | Trophic Level (herbivore, carnivore) |
| Decomposition | Variables were measured per leaf bag |
|  | Weight of leaf litter collected on each sampling date [g] |
|  |  |

1. **Testing your Hypothesis**

You will now examine the data and use statistics to test your hypothesis/es. From your hypothesis/es, you will need to make specific predictions about the data that are provided. Consider the variables that were measured. Which variables relate to your hypothesis/es? What statistics would you use to examine the hypothesis/es?

In **Assignment 3**, you will outline the statistical methods that you will use to test your hypothesis/es. In this assignment, briefly describe how you will use the data provided to evaluate the hypothesis/es you developed in your previous assignment. You should state your hypothesis/es and the predictions that follow from these hypotheses. For each prediction, discuss the statistical test/s that you will use to determine if the data follows a similar pattern to what was predicted and thereby supports the hypothesis. You should specifically discuss what measured variables will be included in your analysis. Table 4 includes a list of variables that are typically used to assess stream conditions that you may find helpful. Also consider the dependent variables that were measured that are listed in Table 3. The more specific you are, the easier it will be to perform your analyses.

Table 4: Benthic macroinvertebrate measures often used when assessing streams (modified excerpt from Barbour et al. (1999)).

|  |  |  |  |
| --- | --- | --- | --- |
| Richness Measures | Composition Measures | Tolerance Measures | Trophic/Habitat Measures |
| No. Total taxa | % EPT | No. Intolerant taxa | % Shredders |
| No. EPT taxa\* | % Ephemeroptera | % Tolerant organisms | % Scrapers |
| No. Ephemeroptera taxa | % Chironomidae |  | % Filterers |
| No. Plecoptera taxa |  |  |  |
| No. Trichoptera taxa |  |  |  |
| Shannon Index |  |  |  |

\* EPT refers to the following insect Orders: Ephemeroptera, Plecoptera and Trichoptera

1. **Reporting your Results and Interpretations**

We are now onto the final and largest assignment. In **Assignment 4**, you will present your findings and discuss your own interpretations of the results. Finally, you will link your findings back to the literature and provide suggestions for future research. The final assignment will be split into 2-3 parts that include the results section, discussion and conclusions. Include figures and tables that display your findings and report the results of the statistical tests. Describe your conclusions and explain how the statistical analyses allowed you to arrive at your conclusions. Do your conclusions support or refute the articles that you read during the development of your hypotheses? Explain why you think this occurred and refer to other literature that supports your argument. This assignment will resemble the results and discussion sections of a scientific paper.

**Literature Cited**

Barbour, M.T., J. Gerritsen, B.D. Snyder, and J.B. Stribling. 1999. Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers: Periphyton, Benthic Macroinvertebrates and Fish, Second Edition. EPA 841-B-99-002. U.S. Environmental Protection Agency; Office of Water; Washington, D.C.

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Ehrenfeld, J.G. 2011. Ecosystem consequences of biological invasions. Annual Review of Ecology, Evolution and Systematics 41:59-80.

Gaire, R., C. Astley, M.K. Upadhyaya, D.R. Clements, and M. Bargen. 2015. The Biology of Canadian Weeds. 154. Himalayan blackberry. Canadian Journal of Plant Science 95:557-570.

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Hall K.J. and H. Schreier. 1996. Urbanization and agricultural intensification in the Lower Fraser River valley: Impacts on water use and quality. GeoJournal 40:135-146.

Invasive Species Council of British Columbia. 2016. Knotweeds. <http://bcinvasives.ca/documents/Knotweeds_TIPS_Final_07_22_2016.pdf>

Leith, R.M. and P.H. Whitfield. 2000. Some effects of urbanization on streamflow records in a small watershed in the lower Fraser Valley, B.C., Northwest Science 74:69–75

LeRoy, C.J. and J.C. Marks. 2006. Litter quality, stream characteristics and litter diversity influence decomposition rates and macroinvertebrates. Freshwater Biology 51:605-617.

Mitchell, R.J., R.S. Babcock, S. Gelinas, L. Nanus, and D.E. Stansey. 2003. Nitrate Distributions and Source Identification in the Abbotsford-Sumas Aquifer, Northwestern Washington State. Journal of Environmental Quality 32:789-800.

**Notes to Faculty**

The primary focus of the series of assignments is to provide students with an opportunity to develop hypotheses based on the literature. We have found that generating hypotheses is an area that many students struggle with. The instructions have been kept deliberately vague, so that students have to direct their own inquiries and to provide faculty with the freedom to tailor assignments to their own specifications. However, faculty should provide students with a brief overview of decomposition in aquatic environments prior to assigning the stream study to ensure that students are comfortable with the following terms: leaf litter, decomposition, benthic macroinvertebrate, and detritivore.

Students benefit from considerable guidance through the assignments. In particular, the hypothesis assignment benefits from the submission of a rough draft followed by a final draft. Faculty may direct students to simpler or more complex hypotheses as they provide formative feedback, thereby tailoring the assignments for each student. Faculty may wish to provide a lecture or laboratory period on basic statistics and/or graphing in excel, or other software. There are many excellent introductions to statistics in Excel available as accompanying resources with textbooks, other TIEE datasets, or internet sites, such as HHMI BioInteractive science education resourses (see <http://www.hhmi.org/biointeractive/spreadsheet-data-analysis-tutorials> or <http://www.hhmi.org/biointeractive/evolution-action-data-analysis>)

Often the hypotheses that students select to examine yield non-significant statistical results. Students struggle with finding no significant effect when they have anticipated one and that they require further direction as to how to interpret a lack of a significant effect. We have found that a non-significant finding provides an opportunity for students to deepen their understanding of the scientific process, and we encourage faculty to avoid pre-selecting only those hypotheses that yield significant results.

It would be beneficial to assign or conduct exercises on basic statistics and spreadsheets prior to Assignment 3. Students can then examine the provided data and propose their analyses. Students with limited experience with statistics or spreadsheets would benefit from the submission of a rough draft, and/or extensive formative feedback prior to the completion of assignment 4.

Alternatively, faculty may wish to select a subset of data to analyze to illustrate particular concepts. Outlined below are areas that yield significant statistical comparisons.

1. **Species Tolerances** – An examination of the relationship between the temperature of each stream site and the abundance of specific orders of macroinvertebrates per leaf bag provides an example of how species tolerances differ. In the faculty dataset, example figures are shown for the significant relationships between Ephemeroptera, Trichoptera and Nematoda and temperature. Students can discuss the direction of the relationships and the magnitude of the slopes of the relationships. Students may be surprised to find no relationship between pH and number of specific invertebrate Orders per bag. In this case, faculty can direct students to examine the range of the pH values and then have students examine the range of pH values tested in the literature. It is also interesting to note that the data was collected in the late summer and fall of 2015. The Fraser Valley region of British Columbia experienced a severe drought during the summer of 2015 which contributed to lower stream flows and higher stream temperatures during the study. The observed relationship between temperature and abundance of different invertebrate groups provides an opportunity to discuss the potential impact of climate change on stream invertebrate communities.
2. **Pollution effects on Biodiversity** – A comparison between site 6 and site 10 provides a dramatic example of the impact of pollution on the community of benthic aquatic macroinvertebrates and on leaf litter decomposition. Site 6 had been impacted by runoff from a dairy farm and the dumping of milk by the diary operators over two decades (see http://www.abbynews.com/news/the-20-year-saga-of-a-fouled-creek/amp/). Site 10 is also situated in a similar rural/urban area but has not had as severe pollution issues. An examination of the invertebrate diversity data clearly shows that high abundance of invertebrates does not necessarily correspond to high biodiversity. One could also compare the leaf litter decomposition at the two sites and theorize about the impact of reduced diversity on energy flow through stream food webs.
3. **Metrics Used for Bioassessment** – Faculty may wish to engage in a more detailed examination of the use of macroinvertebrate data to assess the quality of a stream’s ecological condition than discussed in the previous section. A variety of metrics are used to measure a stream’s condition and the range of conditions present in the dataset provides an opportunity to compare these metrics. Detailed information on biological metrics used to assess streams can be found in Barbour et al. (1999). Below is an excerpt from table 9-1 that includes metrics appropriate to the stream study.

Table 4: Some potential metrics for benthic macroinvertebrates that could be considered for streams (Barbour et al. (1999)).

|  |  |  |  |
| --- | --- | --- | --- |
| Richness Measures | Composition Measures | Tolerance Measures | Trophic/Habitat Measures |
| No. Total taxa | % EPT | No. Intolerant taxa | % Shredders |
| No. EPT taxa\* | % Ephemeroptera | % Tolerant organisms | % Scrapers |
| No. Ephemeroptera taxa | % Chironomidae |  | % Filterers |
| No. Plecoptera taxa |  |  |  |
| No. Trichoptera taxa |  |  |  |
| Shannon Index (H’) |  |  |  |

\* EPT refers to Ephemeroptera, Plecoptera and Trichoptera

1. **Biodiversity Calculations** – Faculty may wish to use the dataset to demonstrate how to calculate diversity indices and to have students examine the relationship between species diversity, richness and abundance. Faculty may have students calculate diversity indices for each stream type and then compare across the stream types. In the faculty dataset, the calculations for the Shannon Diversity Index (H’) for each site are shown on the Biodiversity Calculations spreadsheet. The calculations were done by using simple formulas separately for each site. Students can be directed to calculate H’ values for each site in a similar manner. A detailed description of how to calculate diversity indices is available in the Appendix of Doherty et al. (2011) Using Stream Leaf Packs to Explore Community Assembly TIEE Vol 7). An additional excel sheet provides figures of the biodiversity, macroinvertebrate total abundance and species richness per site.
2. **Temperature Impacts on Biodiversity and EPT taxa**– The dataset shows a negative correlation between temperature and biodiversity. In this analysis, biodiversity was calculated according to the Shannon Index (H’). Further analysis of the number of shredders per leaf litter bag shows a negative relationship with temperature. Students need to recognize that the analysis should include the number of shredders per leaf litter bag rather than the unadjusted number of shredders column. Litter bags were lost at a few sites, therefore the total number of shredders should be divided by the number of litter bags that remained. The number of litter bags that contributed to the shredder count is detailed in the # bags column in the leaf litter dataset. Similar relationships can be examined with the number of EPT taxa, number of sensitive taxa with temperature, etc. Students can then theorize about the impacts of climate change on community and ecosystem processes
3. **Comparison of Leaf Litter Species** – Students may wish to consider how the quality of the leaf litter may influence decomposition by different organisms, and on the role of different decomposing organisms at different stages of the decomposition process. In the faculty dataset, the results of the analysis on the percentage of leaf litter remaining on each collection day are shown. Thimbleberry was found to have decomposed more than the other species by day 7, yet no differences in decomposition were found at later days. To further examine these findings, students may then analyze the abundance of shredder species present at each collection date. Additional data is provided in a separate worksheet in the student dataset on the nutrient composition and toughness (a correlate of lignin content) of the leaf species examined. Students may wish to examine the C:N, C:P and P:N ratios of the leaf litter to further discuss how these factors may contribute to the difference observed at day 7. It is possible that the difference observed at day 7 is due to microbrial decomposition rather than invertebrate detritivores.

1. **Calculating Decomposition Coefficients** - Faculty may use the leaf litter data to calculate decomposition coefficients. Students may select to calculate the decomposition for a specific leaf species at a specific site, or students may select to take the mean across all the sites of the leaf litter remaining. Students would then plot the percent mass remaining relative to the time in the stream and fit a negative exponential curve to the data. The mass loss through time would be expressed as:

Percent Mass remaining = e –kt

Where e = natural logarithm

t = time unit (e.g. days)

k = decomposition coefficient

The decomposition coefficient (k) can be estimated by fitting an exponential model to the data in Excel. Plot the percent mass remaining relative to time and fit a trendline using an exponential model. The decomposition coefficient obtained from the negative exponential equation can be compared across leaf species, or across sites for one leaf species. Faculty may wish to use the data to demonstrate the importance of measuring handling losses and adjusting for these losses. Students placed 4.0 g of each type of leaf litter in each of the litter bags. A set of leaf litter bags were removed immediately after placement in the streams to account for losses during transport. To account for these handling losses, the weight of leaf litter remaining on day 0 after experimental manipulation should be used as the initial weights to calculate percent decomposition. Faculty may wish to have students calculate percent decomposition based on both 4.0 g and on the weights after transport to compare how these adjustments varied among leaf species.

Rubrics that can be used to grade the assignments and assess the outcomes of the exercise are available at [http://tiee.esa.org/resources/Rubrics for the Stream Study Assignments.docx](http://tiee.esa.org/resources/Rubrics%20for%20the%20Stream%20Study%20Assignments.docx).

**Literature Cited**

Barbour, M.T., J. Gerritsen, B.D. Snyder, and J.B. Stribling. 1999. Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers: Periphyton, Benthic Macroinvertebrates and Fish, Second Edition. EPA 841-B-99-002. U.S. Environmental Protection Agency; Office of Water; Washington, D.C.

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**Useful Resources for Aquatic Ecology Methods and Curriculum**

Hauer, F.R. and G.A. Lamberti. 2017. Methods in Stream Ecology: Volume 1 Ecosystem Structure 3rd Edition. Elsevier Inc., London, UK.

Hauer, F.R. and G.A. Lamberti. 2017. Methods in Stream Ecology: Volume 2 Ecosystem Function 3rd Edition. Elsevier Inc., London, UK.

Leaf Pack Network. Stroud Water Research Center. <https://leafpacknetwork.org/>

WOW. 2004. Water on the Web **-** Monitoring Minnesota Lakes on the Internet and Training Water Science Technicians for the Future - A National On-line Curriculum using Advanced Technologies and Real-Time Data.(www.waterontheweb.org). University of Minnesota-Duluth, Duluth, MN 55812.

**Useful Resources for Teaching the Process of Science**

Understanding Science. 2018. University of California Museum of Paleontology. 3 January 2018 <http://www.understandingscience.org>.

**Useful Resources for Teaching Academic Writing**

http://guides.library.cornell.edu/annotatedbibliography

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