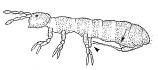
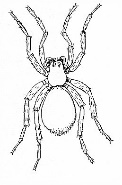
# Image result for soil miteLeaf litter and soil invertebrates

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**Summary:**

The diversity of invertebrates occupying the leaf litter and organic layer of soil can be quantified to examine abiotic and biotic questions. This lab can be used as a starting point for a long-term study or short-term activity to examine invertebrates and how they may influence red-back salamander populations.

**Background:**

Terrestrial salamanders have top-down effects on forest floor ecosystems (Best and Welsh 2014; Hocking and Babbitt 2014). They consume invertebrates, which in turn are responsible for regulating the rate of leaf litter decay. Invertebrates affect many ecosystem functions and are highly responsive to climate change (Prather et al. 2013). Increased temperatures may benefit some litter invertebrates by allowing them to expand their ranges and increase their reproduction rates (Rodenhouse et al. 2009; Ladanyi and Horvath 2010). Alternatively, a reduced snowpack layer could increase mortality rates because of its role as an insulator from freezing temperatures at ground level where these invertebrates overwinter (Bale 1991).

**Supporting documents:** Instructor notes, Excel datasheet, Identification guides (4 documents)

**Goals:**

1. To ask a relevant question and write out hypotheses regarding invertebrate diversity in SPARCnet plots or other locations
2. To collect samples that will be processed to gather data to answer your question
3. To learn techniques of sample collection, Berlese funnel set up and operation, microscope use and invertebrate identification
4. To analyze your data, present your results, determine the level of support for your hypothesis and make conclusions

**Lab steps:**

**Step 1: Determine your question and hypotheses.** Brainstorm questions related to the plots and treatments where you will conduct the study. Instructor could suggest questions to the students or allow them to develop questions on their own. Suggestions include: how do abiotic variables (soil type, understory or overstory cover, etc. ) affect invertebrate populations? , how do biotic variables (density of salamanders or other vertebrates) impact invertebrate populations? Select a question that is interesting to you and then write out hypotheses based on background knowledge of the system you are conducting the study in.

**Step 2: Plan your sample collection and prepare Berlese funnels.** Collect at leastthree leaf litter and soil samples at each plot 1-2 times during the spring and fall each year. Each sample should be acquired using a PVC or metal pipe with a 10 cm diameter. Push the pipe approximately 10 cm into the organic layer of the soil and collect the leaf litter and soil. Place the material in a labeled plastic bag. See alternative sample method below.

The Berlese funnel apparatus should consist of a 75-watt light bulb suspended 12 cm from the top of a Berlese funnel (4 quart; 32 cm diameter). Place a wire 3 mm2 mesh in the funnel to prevent most of the litter from falling through. See separate document on supplies.

**Step 3: Process the samples in Berlese funnels.** Weigh each litter sample to determine initial wet mass. Then place the samples in a Berlese funnel apparatus to capture soil invertebrates in a cup of 70-80% ethanol. Collect invertebrate sample cups after one week in the Berlese funnels and place the remaining litter samples in brown paper bags. Determine dry mass (after Berlese funnel) and final dry mass (after drying in an oven at 50°C for one week) of each sample.

**Step 4: Invertebrate identification and quantification.** Identify invertebrates under a dissecting microscope in a grid-lined (5 mm2) petri dish by examining approximately 5 mL of the sample at a time. The soil invertebrates should be identified to either the class or order level including Acarina, Collembola, Araneae, Hymenoptera, Hemiptera, Pseudoscorpionida, Coleoptera, Diptera, Thysanoptera, Orthoptera, Pauropoda, Symphla, Diplopoda, Chilopoda, Psocoptera, and Diplura, similar to Templer et al. (2012). Use the recommended identification guides for assistance. Record the total number of invertebrates in each class or order in each sample.

If desired, invertebrates can also be classified according to their respective functional food group. Detritivores include Acarina, Collembola, Coleoptera, and Diplopoda. Herbivores consist of both coleopteran and dipteran larvae as well as Hemiptera. Predators include Hymenoptera, Pseudoscorpionida, Araneae, Diptera, and Chilopoda.

**Step 5: Perform analyses and draw conclusions.** Perform statistical analyses as appropriate for your data to examine your question. Discuss your plan for analysis with your instructor before starting. Calculate different measures of diversity to address your question. For instance, you may decide to use the total number of invertebrates, abundance of invertebrates in each of the three functional groups and/or the Shannon-Wiener diversity index. It is a good idea to standardize these response variables by dividing each value by the final dry soil mass of the sample. Create graphs to illustrate your data, assess the results of statistical tests relative to your hypothesis and draw conclusions.

**References (and suggested reading):**

Bale, J. S. 1991. Insects at low temperature: a predictable relationship? Functional Ecology 5:291-298.

Best, M. L. and H. H. Welsh. 2014. The trophic role of a forest salamander: impacts on invertebrates, leaf litter retention, and the humification process. Ecosphere 5:1-19.

Hocking, D. J. and K. J. Babbitt. 2014. Effects of red-backed salamanders on ecosystem functions. PLoS ONE 9:86854.

Ladanyi, M., and L. Horvath. 2010. A review of the potential climate change impact on insect populations – general and agricultural aspects. Applied Ecology and Environmental Research 8:143-152.

Prather, C. M., S. L. Pelini, A. Laws, E. Rivest, M. Woltz, C. P. Bloch, I. D. Toro, C-K. Ho, J. Kominoski, T. A. S. Newbold, S. Parsons and A. Joern. 2013. Invertebrates, ecosystem services and climate change. Biological Reviews 88:327-348.

Rodenhouse N. L., L. M. Christenson, D. Parry, and L. E. Green. 2009. Climate change effects on native fauna of northeastern forests. Canadian Journal of Forest Research 39:249–263.

Templer, P. H., A. F. Schiller, N. W. Fuller, A. M. Socci, J. L. Campbell, J. E. Drake, and T. H. Kunz. 2012. Impact of a reduced winter snowpack on litter arthropod abundance and diversity in a northern hardwood forest ecosystem. Biology and Fertility of Soils 48:413-424.

**Supply list:**

Plastic bags (gallon size)

Sharpies

Galvanized funnels: <https://smile.amazon.com/Stant-75-003-2-Quart-Galvanized-Funnel/dp/B000I182RY/ref=pd_sbs_86_2?_encoding=UTF8&pd_rd_i=B000I182RY&pd_rd_r=71e24340-b69b-11e8-861c-cd06ea1130b1&pd_rd_w=jiq1C&pd_rd_wg=sHD2a&pf_rd_i=desktop-dp-sims&pf_rd_m=ATVPDKIKX0DER&pf_rd_p=53dead45-2b3d-4b73-bafb-fe26a7f14aac&pf_rd_r=WXEXA5KWEPN2RFJWAJ7S&pf_rd_s=desktop-dp-sims&pf_rd_t=40701&psc=1&refRID=WXEXA5KWEPN2RFJWAJ7S>

Metal screening: can be found at hardware stores



Figure 1. Example of a berlese funnel setup with specimen cup for invertebrates below the funnel. Note light bulb suspended above the funnel.