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BIO 181 Lab
Supplemental Learning Aids

Unit 4.1: Leaf Herbivory and Fungal Occurrence – Methods Fall/Sum Online

Ecological Methods Fall/Sum: Field Sampling Techniques

You get to go explore! Enjoy your time outdoors, but be safe!

Instructions: In preparation for data collection, be sure to read the background information in the lab unit pages 47-49, and read all of the steps below. Although you will not be completing this lab exactly as written in the lab manual, we will follow it as much as possible. The methods you will be using are below.

Materials you need on the field:

› **Directions for the Lab Unit/How to collect Data** – bring on paper or be sure you can read on your phone or tablet.
› **Fall/Summer Data Collection Handout** (https://wordpress-projects.wolfware.ncsu.edu/bio-181l-zchxzbn/wp-content/uploads/sites/75/2021/06/HerbivorytsLab-unit_onlineversion_handout-OnlineFallSum.docx)
› a grid for measuring surface area -included in your lab manual (or download a surface area app on your phone such as *LeafByte (https://apps.apple.com/us/app/leafbyte/id1362985339)* for iPhone or *BioLeaf (https://play.google.com/store/apps/details?id=upvision.bioleaf&hl=en_US)* for Android and review the tutorials before going outside),

> a magnifier app for your phone, for example, *Magnifying Glass* for iPhones
  (https://apps.apple.com/us/app/magnifying-glass-flashlight/id908717824)or Android
  (https://play.google.com/store/apps/details?id=com.rvappstudios.magnifyingglass&hl=en_US)
> pen or pencil,
> a clipboard or other writing surface,
> phone camera/regular camera,
> list of random integers (see below, Methods, Step 3)

Safety: Please be mindful of traffic, wildlife, standing water, any sort of brush or vines that may be hazardous. Wear closed-toed shoes and socks to cover your ankles. You may want other protective items like a hat, sunscreen, long-sleeved shirt and long pants.

Methods:

You will be conducting data collection in any area where you can find trees (around your home, neighborhood, a local park, greenway, etc.). NOTE: We are conducting non-destructive sampling, so try not to remove leaves from the trees as you complete data collection.

1- Select an area where you can locate at least three different trees. Ideally, they will be different species of trees so that you can compare them with respect to herbivory and fungal incidence, or you can pick different areas (for example, a completely natural area, a landscaped area, an urban area) and then select the same species of tree in each area. It is up to you how you want to design your study.

> Walk around each tree to visually inspect it, noting any organisms, overall leaf damage, bark
  damage, lichens, mosses, etc.
> Take pictures of each of your trees and any associated organisms or interesting observations
> Identify your trees down to genus and/or species. Use the ID resources in this document.
> Include the location of the tree and its surroundings (add GPS location/geotag, if possible).

Note all of your observations in the space provided in the FallData Collection Handout
(https://wordpress-projects.wolfware.ncsu.edu/bio-181l-zchxzbn/wp-
content/uploads/sites/75/2021/06/HerbivorytsLab-unit_onlineversion_handout-
OnlineFallSum.docx). Also, include a brief description your sampling location(s).

2- Choose one location on the tree that has evidence of herbivore damage and "select" a leaf to evaluate. Look for herbivory and fungal damage/presence using visual analysis (use magnifier app on your phone). Also, refer to images in the Lab Unit ID site (https://wordpress-projects.wolfware.ncsu.edu/bio-181l-zchxzbn/laboratory-units/unit-4-leaf-herbivory-examples/). Without removing the leaf, you will place a 1 cm^2 grid/graph paper transparency over the leaf to calculate approximate leaf surface area. You can also take a picture of the leaf under the clear grid and calculate the leaf surface area later. Alternatively, you may use the surface area app on your phone (see type of approved apps in Materials), and you will have to practice using it before taking your surface area measurements.

Calculations if using a grid: (video (https://mediasite.wolfware.ncsu.edu/online/Play/7ea50a5f709e4c97a8c31145fdd3181c1d) explanation of the process, video (https://mediasite.wolfware.ncsu.edu/online/Play/795f9d8e5b37491e99e6d1bb5a12738b1d) with an actual leaf – view both)

- Full squares or squares more than half full = 1 cm^2
- Half-filled square is = 1/2 cm^2
- Less than half = 0 cm^2
  1. Calculate and record overall leaf area (all possible squares that the intact leaf would take up). NOTE: if a large part of the leaf is missing due to damage, assess what the total area that the leaf would have filled.
  2. Calculate area damaged by herbivory. Take leaf surface area with herbivory taken into account. Count areas that are missing due to herbivory.
  3. This will give you an approximate average surface leaf area lost to herbivores. Record your data in Table 4. 1-1 in the *Data Collection Handout*.
  4. Repeat #2 and 3 looking at fungal infection in each leaf. Record your data in Table 4. 1-1 in the *Data Collection Handout*.

Example: In the image below, the darkened squares represent the part of the leaf still present, and lighter squares represent damage. The total surface area = 14.5 cm^2
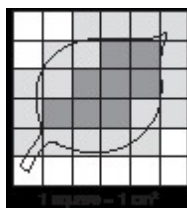
Herbivory leaf surface area = 6.5 cm^2

Figure 4.1-1. Estimation of Leaf Surface Area

3- Repeat this protocol using simple random sampling for a total of 10 different leaves for each of your 3 trees and record your data in Table 4.1-1 in the the *Data Collection Handout*. Use the random integer generator (https://www.random.org/integers/): **Instructions below:

**Random integer generator instructions – Input the following:

Part 1: The Integers

Generate random _10_ integers (maximum 10,000).

Each integer should have a value between _1_ and _10_ (both inclusive; limits ±1,000,000,000).

Format in _2_ column(s).

<u>Part 2</u>: Go! Click on get numbers. Take a picture or screenshot. You will need these in the field.

Using a piece of paper, cut it into four equal pieces and write, "Left," "Right," "Up," or "Down." You will randomly select which way you want to go by shaking up your paper pieces in a bag or other container and choosing one square each time. Return the used square to your bag/container. Repeat until done with all leaves.

<u>Video (https://mediasite.wolfware.ncsu.edu/online/Play/5bbbf3ae37c946c1b33f953d1390d3c21d)</u> of the random sampling technique using the randomly generated numbers and the "Left" "Right" "Up" "Down" squares.

NOTE: If you get "Up" and you cannot go that high or cannot sample that leaf for any reason, go to the next random number until you find a leaf you can sample.

› Take pictures of types of leaf herbivory you observe on your tree.* Insert them into the *Data Collection Handout,* and bring them to the In-Lab meeting.
› Take pictures of types of fungal infection you observe on your tree.* Insert them into the *Data Collection Handout,* and bring them to the In-Lab meeting.
   *(Use the <u>Lab Unit ID site (https://wordpress-projects.wolfware.ncsu.edu/bio-181l-zchxzbn/laboratory-units/unit-4-leaf-herbivory-examples/)</u> and the Resources in the list below to identify the type of leaf herbivory and fungal infection.

4- Be sure to record all of the data in the the <u>Fall Data Collection Handout (https://wordpress-projects.wolfware.ncsu.edu/bio-181l-zchxzbn/wp-content/uploads/sites/75/2021/06/HerbivorytsLab-unit_onlineversion_handout-OnlineFallSum.docx)</u>.

<u>Resources:</u>

Tree Health and Urban Ecology:

13 Steps for evaluating tree health:

<u>https://www.deeproot.com/blog/blog-entries/13-simple-steps-to-evaluate-trees</u> <u>(https://www.deeproot.com/blog/blog-entries/13-simple-steps-to-evaluate-trees)</u>

Damage to landscape trees from NC State's Extension program:

<u>https://gardening.ces.ncsu.edu/weather-2/storm-damaged-landscape-trees/</u> <u>(https://gardening.ces.ncsu.edu/weather-2/storm-damaged-landscape-trees/)</u>

What Tree Should be Planted from NC State's Extension program:

https://lenoir.ces.ncsu.edu/2019/02/what-tree-should-be-planted/ (https://lenoir.ces.ncsu.edu/2019/02/what-tree-should-be-planted/)

Resources for Tree and Organism Identification: Here are some resources in the form of links and applications (some of these apps are not free, but you can search for your own online too):

NC Tree Identification from NC State's Extension program:

https://gardening.ces.ncsu.edu/2015/08/nc-tree-identification/ (https://gardening.ces.ncsu.edu/2015/08/nc-tree-identification/)

Trees – NC State Resources Extension Program:

https://gardening.ces.ncsu.edu/gardening-plants/trees-3/ (https://gardening.ces.ncsu.edu/gardening-plants/trees-3/)

ChopDoc – list of apps for tree identification by leaves and bark:

https://chopdoc.com/how-to-identify-tree-by-leaves/ (https://chopdoc.com/how-to-identify-tree-by-leaves/)

Seek by iNaturalist – app that helps you identify organisms:

https://www.inaturalist.org/pages/seek_app (https://www.inaturalist.org/pages/seek_app)

MyNature Tree Guide:

Android: https://www.amazon.com/MyNature-Inc-Tree-Guide/dp/B00CJ1OYV4 (https://www.amazon.com/MyNature-Inc-Tree-Guide/dp/B00CJ1OYV4)

iPhone http://www.mynatureapps.com/mynature-tree-guide/ (http://www.mynatureapps.com/mynature-tree-guide/)

iBird: http://ibird.com/# (http://ibird.com/)

Peterson's Bird Guide: http://petersonguides.com/apps/apps.php (http://petersonguides.com/apps/apps.php)

Merlin Bird ID: https://merlin.allaboutbirds.org/ (https://merlin.allaboutbirds.org/)

Audubon Bird Guide App: https://www.audubon.org/app (https://www.audubon.org/app)

Butterfly Collection – app for identifying butterflies and other insects:

http://hunter.pairsite.com/butterfly/ (http://hunter.pairsite.com/butterfly/)