Surveying trees for HerbVar

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Why include mature trees?

Mature trees, though harder to study than smaller plants, are a key plant growth-form that could have their own characteristic patterns of interactions with herbivores. Therefore, we want to include enough surveys of mature trees in HerbVar's global sampling to allow us to compare patterns between trees and other growth forms. It is also important to include mature trees because there may be major shifts in tree-herbivore interactions with tree ontogeny, from seedling to sapling and sapling to adult.

We are taking a two-pronged approach to including tree species in HerbVar:

- (1) HerbVar collaborators who do not have a special interest in working with mature trees should restrict their surveys to individuals ≤ 2 m height (seedlings and saplings). That is, survey tree species, but focus on seedlings and saplings. Seedling-sapling surveys won't be representative of all the individuals in a population of a tree species, but these are key stages in tree ontogeny—perhaps the stages in which herbivory is most influential. For seedling-sapling surveys, collaborators should follow the <u>Primary HerbVar Survey</u> <u>Protocol</u> as they would with most other plants. Please note in the metadata that you surveyed only immature individuals (≤ 2 m) at your site.
- (2) Collaborators who want to survey mature trees should follow the Tree Protocol below:

The Tree Protocol

Follow the <u>Primary HerbVar Survey Protocol</u> as much as is feasible to pick a survey site, set up a transect, and randomly select focal trees or quadrats with the following key differences:

- Many tree species cover huge geographic areas, making it unreasonable to survey an entire "population" or define a discrete study "site." If you are working with a widespread species, it is fine to choose a "representative" study area, which actually might just be a small part of a large stand of trees.
- Once you have selected a representative study area, you will need to randomly select 30 trees to survey (plus their nearest conspecific neighbors). There are many ways to do this. Here are three methods in somewhat decreasing order of amount of work and rigor:
 - a. Follow the <u>Primary Protocol</u> exactly, establishing a transect, selecting 30 points randomly (distance along main transect and distance from main transect), and using a circular quadrat at each point to randomly select 1 individual of the tree species within the quadrat (as in the Primary Protocol). Survey each selected tree and its nearest neighbor.
 - b. Follow the <u>Primary Protocol</u> except skip the circular quadrat step, which could need to be prohibitively large in some tree populations: Establish a transect and randomly select 30 points (distance along main transect and distance from main

transect). Then select and survey the individual nearest to each random point (plus nearest neighbor).

- c. For trees that are at low density or low abundance, please consult our <u>Low-density/low-abundance protocol</u> to select trees. For low-abundance plants, we recommend surveying every plant within some area. Take GPS coordinates for each plant. Try to get as close to 60 plants as possible. If you are taking GPS coordinates for each plant, then you do not need to measure distances to nearest neighbors because we can measure spatial relationships using the GPS data.
- d. If your plants are at a high enough density that you can't do all of the individuals in an area and you are unable to use a transect to select plants, then this is one method that may work. Walk through your population. Close your eyes and point. Open your eyes and survey the tree you were pointing at and its nearest neighbor. Record the GPS coords of every focal tree and every neighbor tree. Walk some more and repeat until you have surveyed 30 focals and 30 nearest conspecific neighbors. This method will work relatively well in even-aged stands where all trees are roughly similar in size. This will work less well in stands with large variation in tree size because the method is biased towards selecting larger trees more frequently.
- However you select trees, please make sure to take detailed notes on what you did.
- Note that some of the trees you select may be seedlings or saplings. We recommend doing whichever individual you randomly select, regardless of its age. This should yield a representative sample of all individuals at the site, across age classes.

Record site-level and tree-level data

For each tree, sapling, or seedling, please follow the <u>Primary Protocol</u> as closely as possible to record tree-level data. There are columns for these variables in the HerbVar <u>Datasheet</u> <u>Template</u>.

Site-level data to record include:

- Plant species, genus, family
- Latitude and longitude of transect origin (if using a transect, otherwise collect coordinates for each tree)
- Site diameters (longest diameter and perpendicular diameter)
- Habitat type
- Natural or managed
- Estimated density of focal species within site (e.g., N stems per m²)
- etc.

Tree-level data to record include:

- Tree distance along transect and distance left or right of transect (or tree latitude and longitude if not using a transect)
- Plant stage (seedling, sapling, adult, reproductive)
- Plant size (height or diameter at 1.3 m)

- If feasible, percent cover (birds-eye view) of focal species within each quadrat or within a reasonable radius from each focal plant
- Herbivore (see immediately below)

Quantifying herbivory on trees

- For seedlings and saplings (≤ 2 m height), please follow the Primary Protocol to estimate herbivory at the whole-plant and per-leaf scale. As a reminder, this involves 3 herbivory estimates:
 - $\circ~$ (1) Leaf-level percent herbivory estimates for 10 randomly selected leaves
 - (2) Counts of presence/absence of herbivory for up to 60 leaves per plant
 - (3) A whole-plant visual estimate of herbivory
 - And of course please record the number of galls, mines, and other discrete damage types from sessile herbivores
- For mature trees:
 - Randomly select 30 leaves for quantitative estimates of % herbivory on each of the 30 leaves
 - See <u>Primary Protocol</u> and <u>Damage Estimation Training Document</u> for guidelines on quantifying percent herbivory per leaf
 - Please also record the number of galls, mines, and other discrete damage types. Note that mines should be included both in percent damage (because they represent damaged surface area) and as counts.
 - Randomly select an additional 30 leaves to score for presence/absence of herbivory. Record the number out of 30 with herbivory.
 - See <u>Primary Protocol</u> and <u>Damage Estimation Training Document</u> for guidelines on quantifying presence/absence of herbivory per leaf
 - Do not worry about estimating herbivory at the whole-plant scale for mature trees; we will estimate this using the 30 presence/absence leaves and the 30 percent herbivory leaves

Selecting leaves for herbivory estimates

Trees... are tall. And we will not be able to reach top branches. **We will therefore focus on low branches** that can be reached from the ground with a pole pruner, and sample from multiple places around the circumference (see right).

Randomly choose leaves within the tree. We provide some guidelines below, but you should choose an approach that makes sense for you and your species.

Remember that we are trying to acquire a random subsample of all leaves on the tree; this

means avoiding any preference for/against particular leaves (e.g., young vs old). Ideally, leaves will be sampled in proportion to their frequency on the tree.

- The easiest method, if it would work for your trees, is to close your eyes, point at the tree, open your eyes, and take the leaf you were pointing at ("lan's nose pointing method" in the <u>Damage Estimation Training Document</u>).
- Perhaps the most rigorous but most time-consuming method is to haphazardly strip several times as many leaves as you need (e.g., >200 leaves). Place leaves individually into a large bag. Mix them. Close your eyes and draw 30 leaves for percent herbivory and 30 leaves for presence/absence of herbivory.