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American Beech Tissue Collection for DNA

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Hemlock Landscape Gen...



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We use this protocol and it's working

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Abstract

Steps for collecting tissue from American Beech trees for DNA analysis. The Plant Computational Genomics lab is conducting a landscape genomics study of climate adaptation in the American Beech. This protocol is a step-by-step guide to collect a tissue sample. The protocol records metadata in TreeSnap (<https://treesnap.org>) and is designed to allow revisiting a collection site to collect more data by future researchers.

Introduction

- 1 This protocol is intended for field collection of leaf tissue for the American Beech landscape genomics project sponsored by the **Plant Computation Genomics** lab at the University of Connecticut. The goal of the project is to identify climate adapted genomic variation for seed banking and potential use in future breeding programs or for assisted migration.

A complete data collection will include the sample, metadata of the tree, and photographs. There is no minimum sample size for a collector, but 10 samples from a collector is appreciated.

Collectors will receive a collection kit in the mail. Metadata will be entered into **TreeSnap**. Samples can be mailed to:

Microbial Analysis, Resources, and Services (MARS)
UConn CORE
ESB 309 Unit-3032
67 North Eagleville Rd.
Storrs, CT 06269-3032

Collection Materials

- 2 You will receive in the mail a collection kit with:

- Reusable mailer
- Coin envelopes
- Plastic ziplock bag to hold all samples
- Silica gel packets

Cutting shears or knife, a smart phone, and GPS are required, but not supplied.

Download and install **TreeSnap** onto your smartphone; you will also need to create an account. TreeSnap is a light weight app for collecting metadata for tree studies.

Going into the field

- 3 A few things to remember when you are ready to collect.

1. Always collect with permission. For managed lands, a permit or permission from the land manager is required. For collecting on private lands, always ask the land owner before

collecting. Most people love hemlocks and will give permission to make a small collection.

2. Bring your collecting kit, cutting shears, GPS, and smart phone.

3. Bring a buddy and have fun collecting!

Finding a site

- 4 Road side botany is great fun, but be sure you aren't collecting a planted tree by going down a trail away from where landscaping may be taking place. Only trees of wild provenance will be used in downstream analyses.

Choose a site based on these characteristics:

- You have permission to collect
- Ideally 16-32 km (10-20 miles) distant from any other site
- Accessible by a short walk/hike
- Will make a good TreeSnap entry

Identifying a tree to collect

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- American Beech trees should be adults or established juveniles (>4" DBH).
- Regenerating trees should be sampled only if no adult trees can be found.
- Trees should ideally be accessible for tissue collection by hand or with a pole pruner near a trail head or road.
- Collect trees of wild provenance. In a rural or urban setting, it's impossible to know if a tree near a house was planted. Avoid collecting in someone's yard.

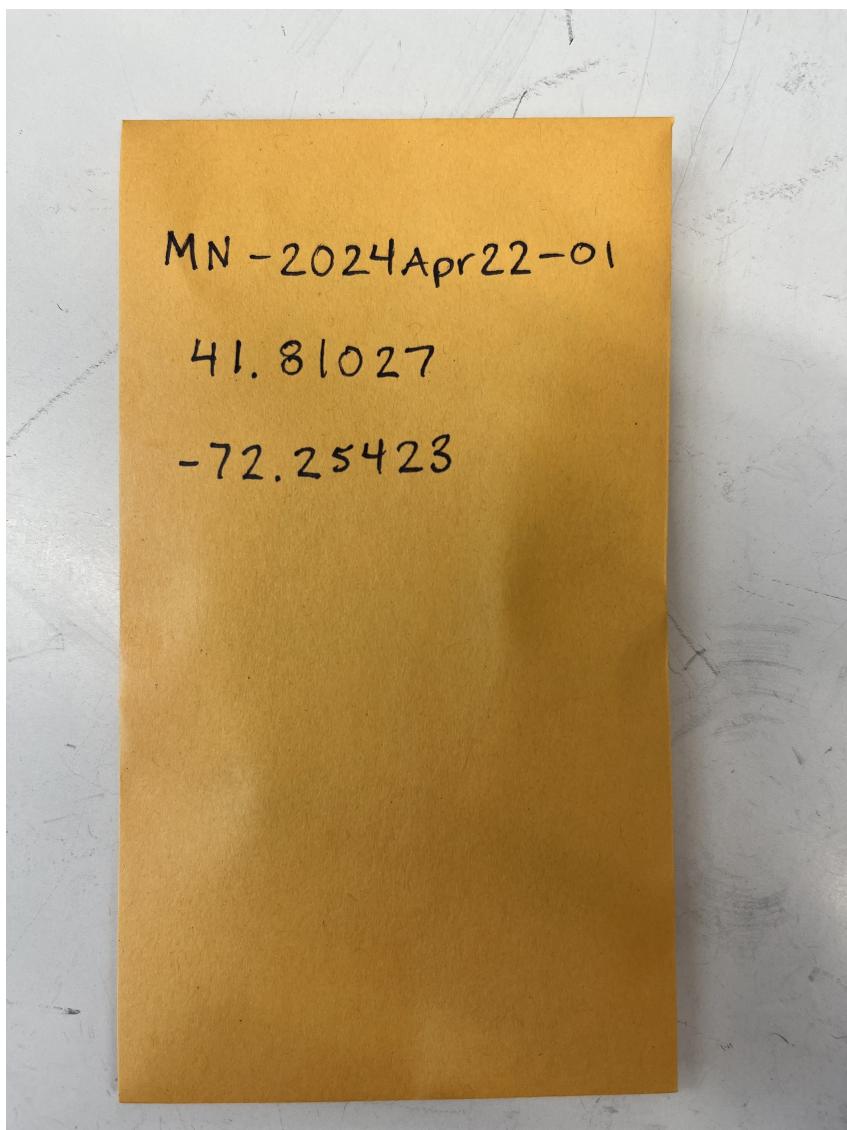
European Beech may co-occur; be sure you know how to [differentiate the species](#).

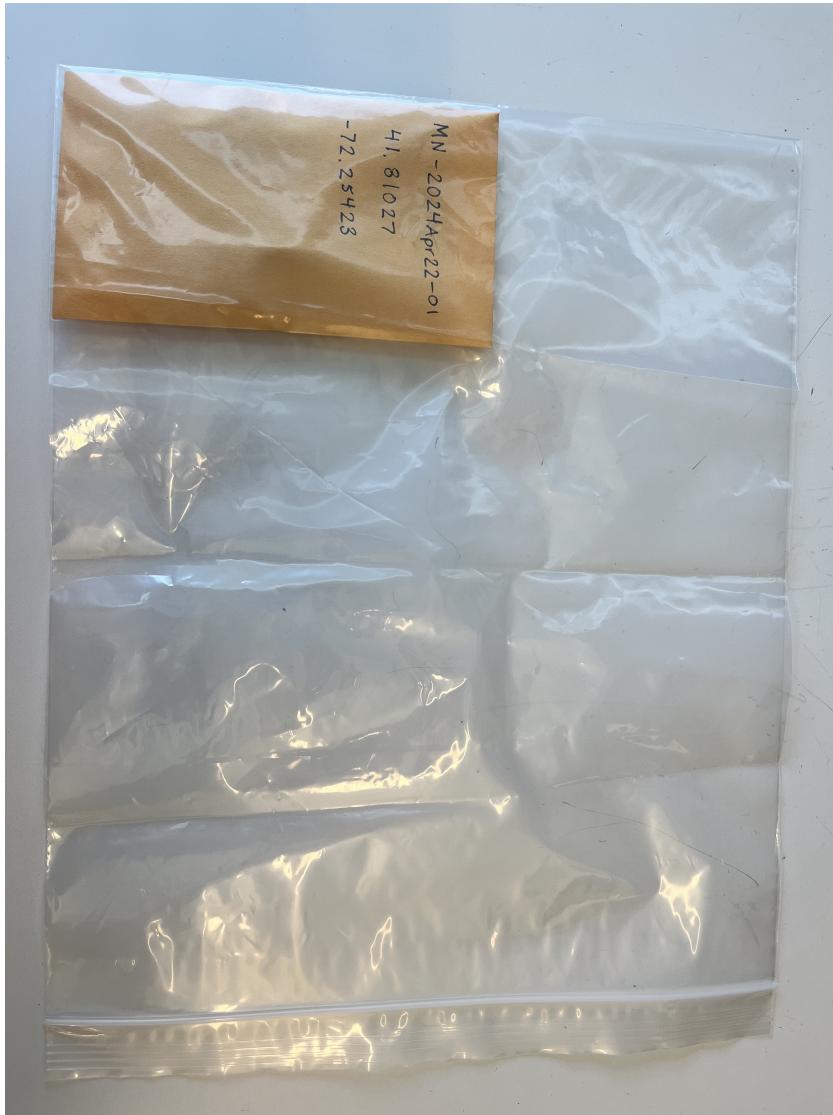
Collecting tissue

6



- Collect 3 healthy leaves from the tree. Make the collection by cutting the stem with a knife, shears, or pinching off the leaves.
- Deposit the leaves into a coin envelope.
- Add 2 silica gel packets.
- Label the envelope with "your initials"- "YearMonthDate"- "CollectionNumber". For example, my first collection for today (April 22, 2024) would be MN-2024Apr22-01.
- Write the latitude and longitude of the tree on the coin envelope (available in TreeSnap).
- Take a picture of the envelope, which will be added in TreeSnap (step 7)
- Place the envelope in the plastic bag.



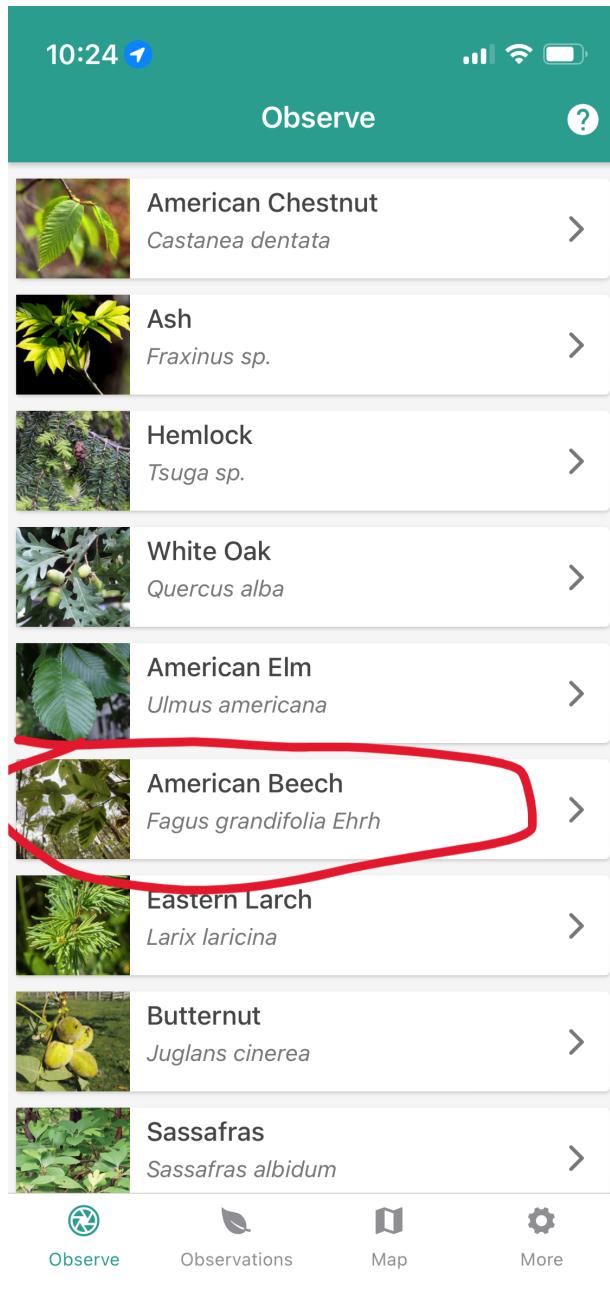


It is essential to write the sample ID and the coordinates on the envelope. If TreeSnap fails, the written data is the backup. Put the sample in the bag with the silica gel. Reseal the bag to dry the sample. Moist samples will mold and the DNA will be degraded.

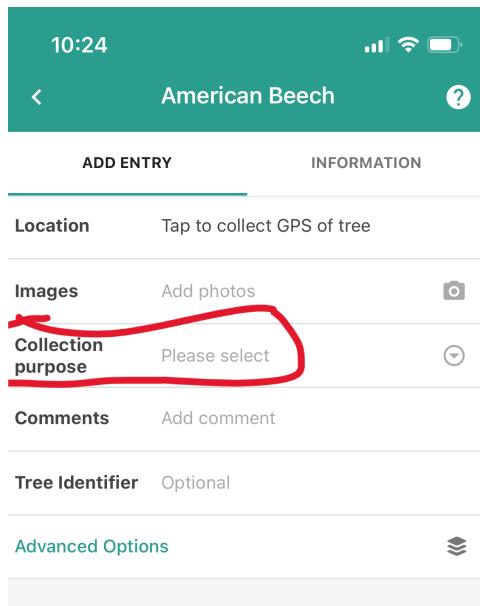
Metadata

- 7 Metadata should be collected which allows 1) revisiting the tree, 2) general health characteristics, and 3) a general description of the site. A photograph should be taken of each tree. Use TreeSnap to collect these data.

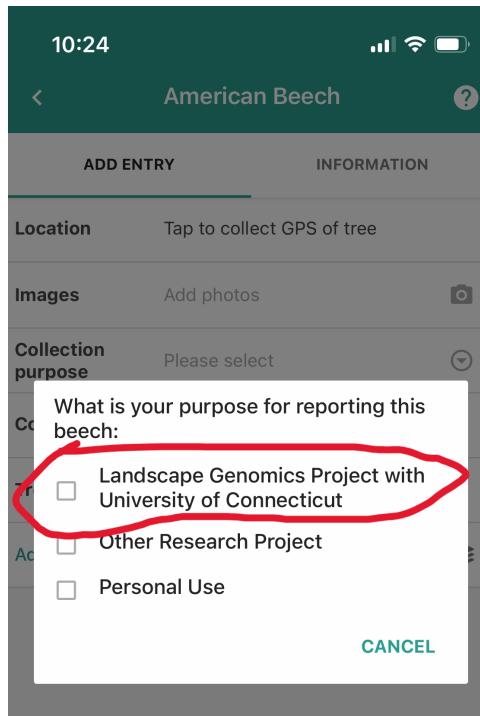
Open TreeSnap and click on the American Beech button.



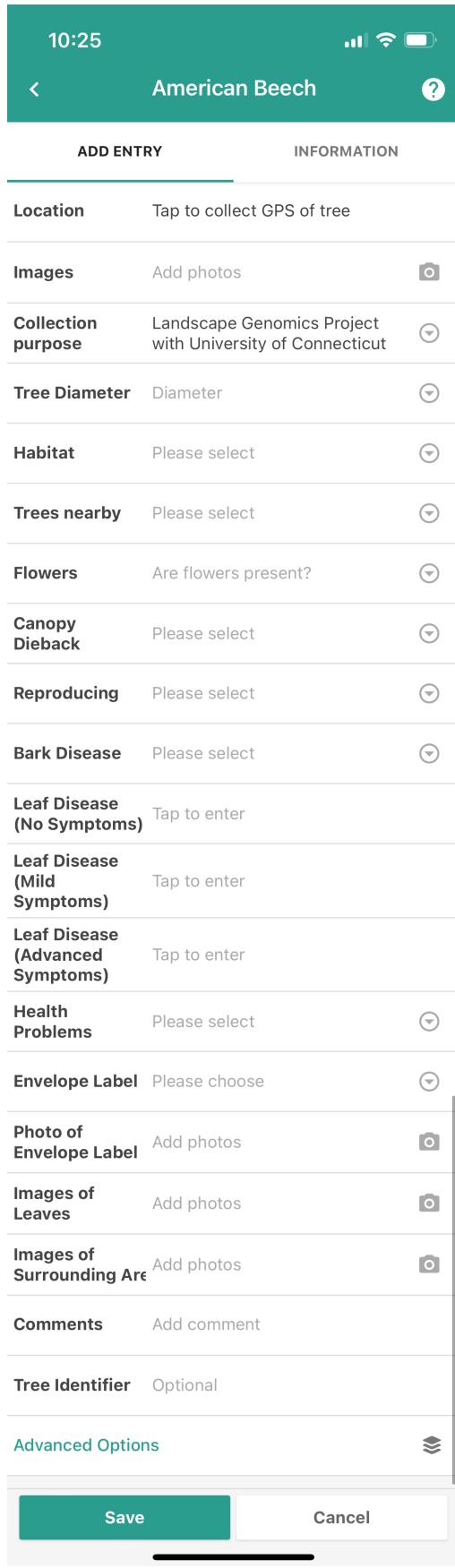
Select "Collection Purpose", then "Landscape Genomics Project with University of Connecticut"



TreeSnap Collection Purpose



Enter information into each field as applicable.



The screenshot shows a mobile application interface for entering data about an American Beech tree. The top bar indicates the time is 10:25 and shows signal, Wi-Fi, and battery status. The title "American Beech" is centered above the form, with a back arrow on the left and a help icon on the right.

The form is divided into two main sections: "ADD ENTRY" (active) and "INFORMATION".

ADD ENTRY

- Location:** Tap to collect GPS of tree
- Images:** Add photos (with camera icon)
- Collection purpose:** Landscape Genomics Project with University of Connecticut (with dropdown icon)
- Tree Diameter:** Diameter (with dropdown icon)
- Habitat:** Please select (with dropdown icon)
- Trees nearby:** Please select (with dropdown icon)
- Flowers:** Are flowers present? (with dropdown icon)
- Canopy Dieback:** Please select (with dropdown icon)
- Reproducing:** Please select (with dropdown icon)
- Bark Disease:** Please select (with dropdown icon)
- Leaf Disease (No Symptoms):** Tap to enter
- Leaf Disease (Mild Symptoms):** Tap to enter
- Leaf Disease (Advanced Symptoms):** Tap to enter
- Health Problems:** Please select (with dropdown icon)
- Envelope Label:** Please choose (with dropdown icon)
- Photo of Envelope Label:** Add photos (with camera icon)
- Images of Leaves:** Add photos (with camera icon)
- Images of Surrounding Area:** Add photos (with camera icon)
- Comments:** Add comment
- Tree Identifier:** Optional
- Advanced Options:** (with gear icon)

INFORMATION

At the bottom, there are "Save" and "Cancel" buttons.

- In the 'Tree Identifier' field, enter the envelope label ("your initials"- "YearMonthDate"- "CollectionNumber").
- In the comments field, add a short description of the site. For example, "Tree located at the Happy Logger trail head", or "Near intersection of Pond St and VT 116".

The lat-longs of the tree are automatically saved when you create the entry (not when you save it!). So be sure to create the entry next to the tree. If you have a GPS that is more precise than your phone and you wish to submit those points instead, enter them in the comments field.

Don't worry if you don't have cell phone reception in the field. The data is temporarily saved on your phone. After you connect to cellular service or wifi again, open the app and select the yellow pop-up box to upload data to TreeSnap.

Save the entry and move on the next tree.

After you've collected

- 8 Be sure your samples are in the resealable ziplock. Put the ziplock in the mailer and reseal it. Using the provided postage, mail the samples to:

Microbial Analysis, Resources, and Services (MARS)
UConn CORE
ESB 309 Unit-3032
67 North Eagleville Rd.
Storrs, CT 06269-3032

Please email notify Michelle Neitzey when samples are mailed. Thank you!