

## Sample Collection Guidelines

Not all forest health problems can be diagnosed in the field. Sometimes it is necessary to collect samples, examine them closely indoors or under magnification, and to compare them with identification keys, insect or disease manuals, and/or online resources. In other cases, it may be necessary to collect and ship samples for identification by a specialist or submit them to a laboratory for diagnosis or assessment. In North Carolina, there are several services available to the public for the diagnosis of plant disorders, insect identification, soil analysis, and plant nutrient analysis.

Samples must be collected and stored properly to be useful for these more detailed investigations. Improper sampling and storage techniques may result in the loss of important specimen parts, may cause deterioration of key characteristics, and/or render the sample unsuitable for laboratory tests. In addition to the sample itself, there is a specific set of information that must be provided along with samples when they are submitted to specialists for assistance. Utilization of the following tips and guidelines will ensure that field samples are collected, stored, and shipped properly.

### General Tips

- 1) ***Fresh samples are better than old ones.*** When plant material is collected in the field, place it into a cooler with ice packs for short term storage. Do not allow the plant to get wet or to dry out. Samples should be shipped as soon as possible after collecting. Avoid shipping at the end of the week; samples that arrive at the end of the week or over the weekend may be significantly deteriorated by the time they are examined.
- 2) ***Living specimens are better than dead samples.*** If possible, insects (especially larvae) can be collected and shipped alive. If the insect is feeding on a plant, include some plant material in the shipment to increase its chances of survival. Plants that can be shipped with root systems and surrounding soil in tact can survive for several days. Microorganisms are more likely to survive until they can be examined in the laboratory if they are kept cold (not frozen).
- 3) ***Collect as much as possible.*** For plant problems, send several specimens including dead, dying, and even healthy examples. Small plants can be dug up and sent with roots and soil attached. Do not remove foliage from a tree; instead, collect symptomatic twigs or branches with foliage attached. Roots (preferably with soil) should be sent whenever possible. For insects, collect several specimens and all available life stages when possible, and include infested plant material when appropriate.
- 4) ***Keep soil and water off of foliage.*** For small plants collected in their entirety, wrap the root ball and attached soil in a plastic bag tied off tightly at the bottom of the stem. If the soil is dry, mist or water lightly to keep the plant from drying out. Excess water should be drained or blotted off. Wrap foliage in newspaper or paper towel to keep dry. Do not seal plants in air tight plastic bags because excess moisture can build up; loosely tied paper or plastic bags are preferred.
- 5) ***Be aware of quarantines.*** It may be illegal to ship certain species of plants, insects, or microorganisms out of quarantined areas. Quarantines may also prohibit the shipment of soil out of areas infested with certain species of noxious weeds. Quarantines are usually established by the state or federal government. Be aware of quarantines established in the county the collection is made, and do not ship quarantined materials out of the quarantine zone. Lists of internal quarantines currently established in North Carolina are available from the North Carolina Department of Agriculture (NCDCA) and/or the U.S. Department of Agriculture - Animal and Plant Health Inspection Service (USDA APHIS).
- 6) ***Collect as much information as possible.*** Collection of the sample itself is only the first step. When submitting a sample, provide as much information as possible. A detailed write-up or list should be included with every sample with the following information, and any other information you feel may be important:

Collector's Name	Description of Signs
Date Collected	Description of the Site (and map)
Location Collected	Recent Pesticide/Chemical Applications
Landowner/Homeowner Info	Recent Activities (e.g. Pruning, Irrigation, Fertilization, Thinning)
Plant Species	Recent Disturbances
Plant Age	Number of Plants Affected
Description of Submitted Sample	Size of Area Affected
Description of Symptoms	Pictures of Plant and Surrounding Area
Timeline of Symptom Development	

### **Small Plants**

- 1) Select a number of plants with a range of symptoms. Include a range of healthy, symptomatic, and recently killed plants.
- 2) It is important to select symptomatic plants at the edges of the area being affected.
- 3) Dig up the entire plant. Keep as much of the root system and surrounding soil intact. Do not pull plants from the ground.
- 4) Mist or lightly water the root system if it is excessively dry. Excess water should be drained or blotted off.
- 5) Place plant in a pot if possible. Wrap the soil and roots in a plastic bag tied off tightly at the lower stem.
- 6) Wrap foliage lightly in newspaper or paper towels.
- 7) Place the entire plant in a loosely sealed plastic or paper bag.
- 8) Ship as soon as possible using the shipping guidelines described below.

### **Foliage Diseases**

- 1) Select several small twigs or branches with foliage attached. Include examples of twigs/branches with a range of healthy, symptomatic, and/or recently killed foliage.
- 2) Strip away bark from several affected twigs or branches that will not be shipped and examine the sapwood for discoloration or streaking; if observed, include larger branch or stem samples with vascular symptoms (*see vascular samples*).
- 3) Cut branches or twigs with a sharp, by-pass type pruning shear or lopper. Do not tear or break off twigs and branches.
- 4) Cut branches to length (if necessary). If the sample is wet, blot it dry. Wrap foliage lightly in newspaper or paper towels and seal in a plastic bag.
- 5) Place sample in a cooler with ice or ice packs. Store in a cool dry place.
- 6) Ship as soon as possible using the shipping guidelines described below.

### **Vascular Diseases**

- 1) Select several small branches that are wilting, discolored or dying (do not select dead branches).
- 2) Strip away bark from affected branches and examine the sapwood for discoloration or streaking; if observed, include branch samples with vascular symptoms.
- 3) Cut branches with a sharp, by-pass type pruning shear or lopper. Do not tear or break off branches.
- 4) If the entire tree is affected and expected to die, examine the sapwood of the main stem. If discoloration or streaking in the xylem is observed, removed a large chunk of the sapwood (preferably with bark attached) using a saw or ax.
- 5) Seal branch and/or stem samples in a plastic bag. Do not allow samples to dry out.
- 6) Place sample in a cooler with ice or ice packs. Store in a cool dry place.
- 7) Sterilize the cutting edge of the tools used to take the sample using alcohol or bleach before using again.
- 8) Ship as soon as possible using the shipping guidelines described below.

### **Cankers, Galls, Swellings, and Weeping**

- 1) Symptomatic branches should be cut with a sharp, by-pass type pruning shear or lopper. Do not tear or break off branches.
- 2) Symptomatic areas on the main stem should be cut out or the entire stem section removed using a saw.
- 3) Seal branch and/or stem samples in a plastic bag. Do not allow samples to dry out.
- 4) Place sample in a cooler with ice or ice packs. Store in a cool dry place.
- 5) Sterilize the cutting edge of the tools used to take the sample using alcohol or bleach before using again.
- 6) Ship as soon as possible using the shipping guidelines described below.

**Large Trees**

- 1) Because large trees cannot be submitted, pictures of the entire tree, close-ups of symptoms, and the surrounding area should be submitted along with samples.
- 2) Collect symptomatic foliage and branches as described above.
- 3) If the tree is expected to die (or has recently died) collect a section of the main stem near the soil line, or if possible, the entire root crown region (lower stem and upper roots). Seal in a large plastic bag.
- 4) Collect root samples if possible, including large buttress roots if the tree is expected to die. Seal in a plastic bag.
- 5) Collect a large soil sample containing fine roots. Seal in a plastic bag.
- 6) Place samples in a cooler with ice or ice packs. Store in a cool dry place.
- 7) Sterilize the cutting edge of the tools used to take samples using alcohol or bleach before using again.
- 8) Ship as soon as possible using the shipping guidelines described below.

**Mushrooms, Conks, and other Fruiting Bodies**

- 1) Hard, perennial conks can be broken off the tree and stored in a loosely sealed plastic or paper bag.
- 2) Mushrooms and other soft fruiting bodies should be blotted dry and sealed in a plastic bag.
- 3) If possible, collect mushrooms or conks with surrounding soil or plant tissue attached.
- 4) For mushrooms, take note of the plant species in the surrounding area.
- 5) Place samples in a cooler with ice or ice packs. Store in a cool dry place.
- 6) Ship as soon as possible using the shipping guidelines described below.

**Insects**

- 1) Most adult insects can be collected and placed immediately into a vial containing 70 percent alcohol.
- 2) Sapsucking insects such as mites, scales, aphids, thrips, and adelgids should be submitted alive on infested branch, twig, or foliage samples (*as described above*).
- 3) Adult butterflies and moths can be collected and killed in a container containing ethyl acetate or placed in a freezer; package lightly in soft tissue paper inside a solid container. Placing moths or butterflies in alcohol will destroy them.
- 4) Larvae, when possible, should be submitted in or on the plant tissues upon which they are feeding in a sealed plastic bag. Otherwise, they should be placed in a vial of 70 percent alcohol.
- 5) Grubs can be dug up and submitted with several cups of soil sealed in a plastic bag.
- 6) Place samples in a cooler with ice or ice packs. Store in a cool dry place.
- 7) Do not allow insects to overheat; they will soften and begin to decompose rapidly.
- 8) Ship as soon as possible using the shipping guidelines described below.

**Shipping Guidelines**

- 1) Mail or deliver samples as soon as possible after collecting. Avoid shipping at the end of the week; samples that arrive at the end of the week or over the weekend may be significantly deteriorated by the time they are examined.
- 2) Soft plant tissues, mushrooms, vascular tissue, samples containing microorganisms, and living insects should be shipped overnight/next day when possible.
- 3) Avoid shipping wet samples; drain or blot excess water. Do not seal samples in plastic bags for more than a few days.
- 4) Ship samples in a Styrofoam cooler containing ice packs sealed in a plastic bag (in case of leakage) when possible.
- 5) Pack samples in a sturdy box adequately filled with packing material such as newspaper. Do not crush samples.
- 6) Seal any liquid containing items (ice packs, vials, wet soil) in a leak-proof plastic bag or container.
- 7) Include all relevant documentation and pictures in a sealed plastic bag.

