

# Sampling Instructions for Nematode Assays<sup>1</sup>

Tesfamariam Mekete and William T. Crow<sup>2</sup>

Nematodes are thread-like microscopic worms that live in all habitats, especially soil and water. Most species are beneficial, but some are harmful parasites of plants and animals. Plant-parasitic nematodes are characterized by having a needle-like structure in the mouth called a stylet that, depending on the nematode species, is used to feed on plant parts that may include roots, stems, leaves and seeds. While some nematode problems can be accurately diagnosed in the field, laboratory assay of soil and roots is usually necessary to confirm field observations. Please follow these instructions carefully for collecting and submitting a sample to the Nematode Assay Lab so that it reaches us in good condition, and you get accurate information from it. Remember, the accuracy of the results is directly related to the quality of the sample.

## 1. Sample Correctly

The sample should be prepared from a mixture of 10 to 20 “cores” of soil. Cores are most easily taken with a soil sampling tube, auger, shovel, or trowel. Use a shovel to cut a 1-inch-thick slice of soil through the soil profile, and then collect a 1- to 2-inch vertical band from the slice. It is often best to discard the top 1 inch from each core since nematode numbers may be very low there.

## 2. Sample at the Right Time and Place

The timing of nematode sampling is important because nematode populations fluctuate throughout the year. Nematodes may be undetectable during the winter and early spring but often increase to a very high population density in the early fall before crop harvest when living roots are present. After harvest, nematode populations may decline sharply. Thus, taking samples at the appropriate time for a crop decreases the risk of failing to detect a damaging nematode species and helps avoid or reduce the potential of nematode problems in the future. Sampling in the early fall facilitates decision making in implementing appropriate nematode management practices before the next crop is planted.

Because nematodes feed on plant roots, always sample from among roots of the plants for which diagnosis is needed. Sample only when soil moisture is appropriate for working the field; avoid extremely dry or wet soil conditions. For the safety of lab personnel who handle the soil, ***do not sample for at least 4 weeks after a nematicide has been applied to give the nematicide sufficient time to dissipate.***

Specific directions for collecting samples from different types of symptomatic plants are as follows:

**Annual Crops** (most vegetables, annual ornamentals, and field crops)—Take soil from root zones of 10–20 affected

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plants that are not yet dead. Include “feeder” or fine roots from several of them. Remove the surface inch of soil before taking each core 6–8 inches deep.

**Fruit and Nut Trees, Perennial Shrubs and Trees**—If many plants are affected, include cores from several of them in the sample. If only one or a few are affected, take several cores from around each plant. Use a spade or shovel to dig within the “drip-line” (the area covered by the branches) to find fine feeder roots. Each “core” should consist of a few fine roots and soil from immediately around them. Discard roots and soil from the surface inch. On most trees and shrubs, cores 8 inches deep should be sufficient. However, for burrowing nematode of citrus, collect roots and soil from below 12 inches deep.

**Turfgrasses**—Collect 10 to 20 cores from areas of declining but not yet dead turf. Collect cores 3–4 inches deep near the desired plant species, avoiding bare spots and weeds. The sample must consist of mostly soil with a few roots; discard foliage.

Advisory or predictive samples are taken to predict the risk of nematode injury to a crop to be planted, or later on in the season for turfgrasses and other perennial crops. For annual crops, nematodes are most numerous and easiest to detect near the end of a growing season. Therefore, sample results are generally more meaningful when the samples are collected immediately following the previous season’s crop rather than immediately preceding the crop to be planted. Predictive samples should be representative of the entire sampling area. Collect cores in a regular pattern over the area. One sample should represent no more than 10 acres for relatively low-value crops such as corn and soybeans, and no more than 5 acres for higher valued crops such as vegetables and tobacco. Areas that have different soil types or that were planted to different crops (or varieties) during the past season should be sampled separately.

### 3. Place All Soil Cores from the Sampled Area into a Plastic Bag

If more than one area is to be sampled, use different plastic bags for each sampling area. The approximate soil volume for a properly collected sample should be 1 to 2 pints. Include as many fine roots as possible (up to ½ cup) mixed in with the soil sample. A 2-pint sample in the bag should fit in a small box like those provided at United States Postal Service offices. Seal the sample in the bag to reduce moisture loss. The soil should be handled carefully because rough handling will crush nematodes living among soil particles.

### 4. Label Each Plastic Bag

Provide a sample number or other identification of your own on each bag, and fill out the Nematode Assay soil submission form so that the lab cannot confuse your sample with that of someone else. Label the plastic bags by writing directly on them with a permanent black felt-tip marker or with a permanent pen or pencil.

### 5. Handle and Submit the Sample Correctly

Nematodes will die from overheating, freezing, or drying. Do not leave samples exposed to sunlight or carry them in a hot car trunk or on the dashboard. Do not add water to the sample, even if it seems dry to you. Just package and send it so that the lab will get it in the same condition as when you collected it. If nematodes are killed in handling, they cannot be recovered in the laboratory, and you will receive false results.

### 6. Complete the Nematode Assay Form

The Nematode Assay Form is available here: <http://nematology.ifas.ufl.edu/assaylab/>. Complete the form with all requested information, being sure to print or write clearly. Also ensure that your sample identification on the bag and the information on the form are correctly matched. Complete information about cropping history and plans, symptoms, etc., will help us make a more accurate diagnosis and recommendation. Accurate identification of the plant species (and variety, if possible) for which a diagnosis is needed is absolutely necessary to make a recommendation.

### 7. Send Proper Payment

As of July 1, 2006, the fee is \$20 for each sample from Florida and \$25 for each sample from outside of Florida. We accept checks, money orders, or cash. Make check or money order payable to “**University of Florida.**” Please be sure that the name that appears in the sample identification information on the sample bag and on the assay form also appears on the check.

## 8. Mail, Ship, or Deliver Samples

Get your samples to the Nematode Assay Laboratory as quickly as possible. Our mailing address is:

Nematode Assay Lab  
Att: Dr. Tesfamariam Mengistu  
Building 970, Natural Area Drive  
Gainesville, Florida 32611.  
Telephone (352) 392-1994  
Fax (352) 392-0190.

Please deliver samples Monday through Friday between 8:00 a.m. and 5:00 p.m.

**Services:** We will determine the quantity of each kind of plant-parasitic nematode recovered from the sample. These results and the appropriate recommendations will be written on the Nematode Assay Form. You and your county Extension agent will each be sent a copy of the assay form no later than 10 working days after we receive the sample in the laboratory. In some instances, identifying nematodes to species is necessary to select the best management program. If requested, we will use molecular tools to identify nematodes to the species level. This may require 3-4 weeks additional time. When such a delay is required, the normal report will be returned within 10 days after sample receipt; a supplementary report will be provided when final results are available.

## 9. Stay Informed

For information concerning collecting and submission of samples or the status of samples already submitted, contact Dr. Tesfamariam Mengistu via email ([tmekete@ufl.edu](mailto:tmekete@ufl.edu)) or phone (352-273-3936)