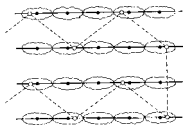
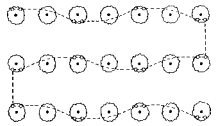


Sampling Procedure to Diagnose Nematode Infestations

Saad L. Hafez



Introduction

Nematodes are minute, worm-like animals that may damage plants but are often difficult or impossible to detect with the unaided eye and are sometimes called the "unseen enemy." The word "nematode" when literally translated means "thread-like." Other names commonly used include "roundworm," "eelworm," or "nema."

Plant parasitic nematodes require the presence of living plants for reproduction and long-term survival. No plant or plant part is totally free from nematode attack, and annual economic losses in the United States have been estimated at 10 percent of the total agricultural crop values. Worldwide, over 2,000 plant parasitic nematode species have been identified and they occupy every available niche provided by plants. When nematodes feed on plant roots the uptake of water and nutrients is reduced, debilitating the entire plant.

Nematode sampling has become increasingly important in modern agriculture as the concepts of Integrated Pest Management (IPM) and Integrated Crop Production are developed and utilized. Scientists concerned with nematode populations have improved methods for their assay; however, data from the best extraction methods are of limited value if the sample is not representative of the area. Effective diagnostic sampling may involve rating plant roots (e.g., galls caused by *Meloidogyne* spp.), bioassays, or visually assessing above-ground growth for effects of foliar pathogens in addition to collecting soil and root samples for nematode counts.

Objectives

Primary objectives of nematode sampling include: diagnosis of disease problems, general detection and surveys; providing advice in IPM programs; and fulfilling research needs, e.g., evaluating different management practices.

Diagnosis

Sampling for nematodes is an increasingly important component of plant disease diagnosis, especially for high value crops and nursery stock. Without confirmation through sampling, poor plant growth because of nematodes may be misinterpreted as nutrient deficiencies or other maladies. The art and practice of collecting representative root and/or foliage tissue as well as soil samples is a critical component of diagnostic sampling. For example, sampling from adjacent, healthy-appearing plants may be just as important as collecting samples from the most severely affected ones.

Detection and survey

Nematode sampling is the basis for determining the occurrence and distribution of many plant parasitic nematodes. Quarantine or phytosanitary regulations of many countries, or political subunits, require that planting materials be produced on land certified free from nematodes. Soil sampling for certification of widely distributed planting materials requires extreme precision for detection of quarantined pests. Although the objective of detection seems simple, a negative result does not necessarily prove absence of the pest, but only indicates that a nematode population is below the detection level.

Advisory

The fact that initial numbers of nematodes can be related to the yield of annual crops has enabled nematologists to develop functional advisory programs, even though relationships between nematode numbers and crop damage may be modified by environment. Because of the importance of reliable detection, most sampling for advisory purposes is conducted when population densities are near their maximum levels, often at the end of the growing season after harvest. Sampling at the time of planting, however, theoretically will give a better estimate of the initial nematode problem where population levels are high enough for detection. Follow-up sampling may be

necessary with perennials because low, nondetectable populations sometimes increase over time to damaging levels.

Research needs

Research is conducted to evaluate the effectiveness of different management practices in reducing nematode populations and plant damage. Extra effort in sampling plant tissues and soil is required to obtain accurate results. The accuracy in determining relative numbers and developmental stages of nematodes may be greatly affected by sample handling or extraction. Design and management for sampling may need to be modified for each specific type of study.

Considerations in designing sampling procedures

1. Nematode distribution patterns will influence the results and must be considered.
2. The capacity of the nematode species to move or be moved by man or other carriers.
3. A majority of nematodes in most annual cropping systems are found in the upper layer of soil (to 15 inches).
4. The influence of biology, feeding habits, and environmental interactions of the nematode species involved.
5. The effect of crop rotation and cultural practices.

How to sample for nematodes

Making the proper management decision for a nematode problem depends on the correct diagnosis, which also depends on proper sample collection, handling, labeling, packaging and shipment. Population densities of plant parasitic nematodes vary greatly in time and space. Population densities are often affected by climate, crops grown, weed hosts, chemicals used, and other factors that may lead to patchy distributions. Patchy or aggregated nematode distributions and changes in the species composition of the nematode communities over time pose major sampling problems. Collection of representative soil samples is the primary component of sampling for diagnosis.

The following recommendations are presented as general guidelines and may require modification for specific field situations.

Time of sampling

a. Sampling should be done before any treatment or management decision is made and before planting.
b. Sampling should occur when nematodes are active and at high populations.
c. Soil should be moist but not excessively wet or frozen.
d. If nematicides are applied to manage a known nematode problem, samples should be taken following application but before planting to determine the effectiveness of treatment.
e. The best time for nematode sampling to make management decisions is usually before harvest (early fall) in preparation for the following season crops, especially sugarbeets or potatoes.

Field mapping

The distribution of nematodes is seldom uniform or constant and changes may occur rapidly. Most of the time nematode distribution is patchy. For these reasons, the field to be sampled should be mapped into subdivisions. Any observable variation in previous crop growth, soil texture, moisture and draining patterns, or cropping history will constitute a subdivision. An effective sampling map may then be constructed.

Sampling Pattern

The sampling pattern is based on the sampling map and should be designed to obtain a reliable representative sample with as little sampling error as possible. An example of a recommended soil sampling pattern for fallow ground or an established crop is shown in Figure 1.

Number, size, and cost of samples

Sample size depends on the area and crop being sampled, the crop's value and any supplementary information to be gathered during sampling. Sample size is measured in terms of the number of cores of soil constituting the sample. The precision of the resulting nematode estimates can be improved by increasing the number of soil cores in the sample. This is also less expensive than increasing the actual number of samples. A useful rule-of-thumb for estimating cost is to consider allocating 1 percent of the crop's total production expense on nematode sampling.

A good approach in sampling is to bulk soil cores in a clean bucket, mix thoroughly, and submit one quart of the mixture for processing. One sample may be used to represent up to five acres of a uniform field subdivision as determined by mapping. Each sample should contain at least 20 individual cores, or if representing less than five acres, at least four cores per acre. Place the sample

in a sturdy, moisture-retaining bag and clearly identify with a tag attached to the outside of the bag. Tags or labels inside the bags may get wet and discolor easily.

Where to take samples

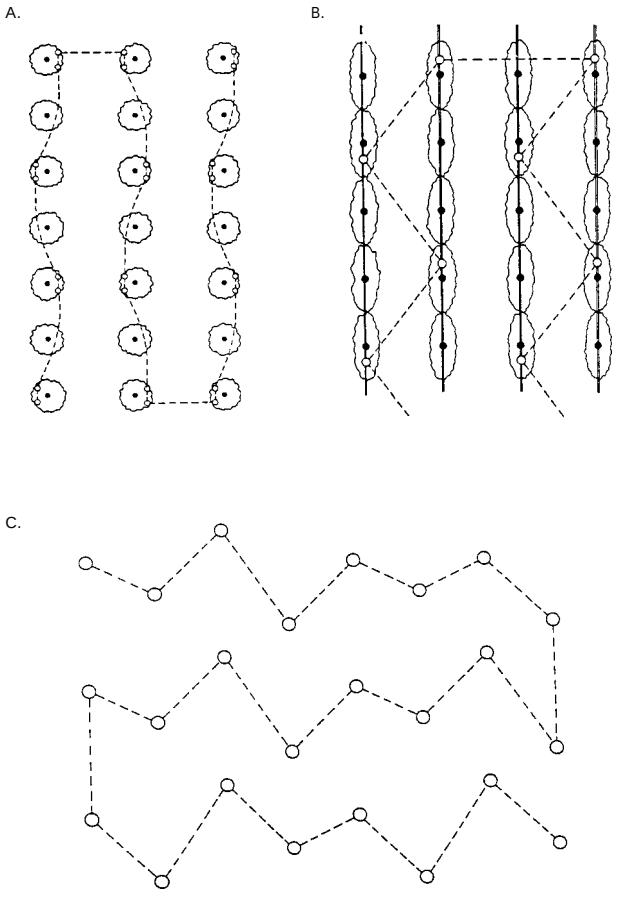
Soil samples should be taken from the plant root zone. If the crop has not been planted, take samples to fit the intended crop's root zone. Plant samples should be taken from the plant parts showing symptoms, if present. Soil or plant samples should be taken from problem areas showing symptoms and from unaffected areas for comparison.

Before planting annual crops, sample cores from a fallow field should be taken by first removing the upper two inches of soil, and then sampling to a depth of 15 to 20 inches. Deeper cores, up to 30 inches, should be taken after prolonged fallow, dry or freezing conditions.

Sampling equipment

Equipment for collecting soil samples for nematode assays includes shovels, soil augers or tubes, and motorized samplers. The typical cylindrical tube sampler designed by soil scientists serves as the standard equipment. Representative sampling tools are shown in Figure 2.

Figure 1



Background information

Include name, location, soil type and texture, observable symptoms (yellowing, necrosis, root rotting, galling, wilting, etc.), cropping history (past several years, current, anticipated), and date of last treatment with a nematicide. This information is invaluable for diagnosis and identification of nematode problems. Refer to the Nematode Diagnostic Laboratory Check Sheet (Figure 3, page 4) for examples of pertinent information.

Storage and delivery

Even the most carefully taken samples may yield inferior results if not stored and delivered properly. Keep the sample <u>cool</u>, ideally at 50 to 55° F. Do not leave the sample in direct sunlight, car trunk or other areas that may heat excessively. An insulated cooler is convenient for sample protection. Deliver or mail the sample immediately to the processing laboratory. Use First Class, UPS, Greyhound or other express delivery and pack well in a sturdy cardboard box or coffee can. *Remember*—Samples must be clearly labeled and accompanied by complete background information. Diagnostic services are available at the University of Idaho, Parma Research and Extension Center, Parma, ID 83660. Telephone: (208) 722-6701.

Figure 2

Figure 1, left: Recommended soil sampling patterns. A. and B., Patterns for perennial plants; C., Pattern for annual crop or fallow field.

Figure 2, above: Examples of soil sampling tools to diagnose nematode infestations.

Figure 3, page 4: Sample form used by the University of Idaho Nematology Laboratory, "Nematode Diagnostic Laboratory Check Sheet."

Figure 3

NEMATODE DIAGNOSTIC LABORATORY CHECK SHEET

Nematode — 061-Y160

Clinic # (to be filled in by clinic)

Requested Analysis and Cost:

O Soil (Cyst not included) $20.00 O Soil & Root: $35.00 O R.K.Identification of Species
O Root or Seed $20.00 O Complete Soil Test: $30.00 Additional $10/species
(Cyst included)

Date Collected _____ Location_____
(Town) (County) (Farm)

Grower _____ Submitted by _____

(Mailing Address) (Phone)

Field Identity _____ # Acres per Sample _____ Sample Type: Soil __ Root __

Crop History: _____ _____ _____ _____ _____
Present or Intended Last Year 2 Years Ago 3 Years Ago 4 Years Ago

Previous Nematode Occurrence _____

Current Soil Treatment: Fumigant or other _____ Date_____
Rate/Acre _____ Application Method _____
(chisel, plow, blade, etc.)

Nematodes Present: Per 500 cc Soil (Approximately 1 pint) Clinic Sample #

Common Name	**/Genus**						Remarks
Northern R.K.	*Meloidogyne hapla*						
Columbia R.K.	*chitwoodi*						
Root-Lesion	*Pratylenchus*						
Stubby Root	*Trichodorus*						
Stunt	*Tylenchorhync*						
Spiral	*Helicotylenchus*						
Pin	*Paratylenchus*						
Dagger	*Xiphinema*						
Stem	*Ditylenchus* spp.						
Ring Nema.	*Criconemella* spp.						
Sheath	*Hemicycliphora* spp.						
Other							
Cyst	*Heterodera*						
Viable							
Empty							
Larvae							
Eggs							

Date Completed _____ _____

Dr. Saad L. Hafez Nematode Lab Director

Phone 208/722-6701 FAX 208/722-6708

NOTE: FAILURE TO DETECT A NEMATODE SPECIES IN A SAMPLE IS NOT PROOF FIELD IS FREE. NEMACH7A\LEF\HAFEZ4

University of Idaho Extension

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