



**United States  
Department of  
Agriculture**

Marketing and  
Regulatory  
Programs

Animal and  
Plant Health  
Inspection  
Service

Plant Protection  
and Quarantine

INTERIM EDITION

# Golden Nematode Program Manual





# Contents

## List of Figures

*page iii*

## List of Tables

*page vii*

## Introduction

Manual *page 1-1-1*

## Procedures

Introduction *page 2-1-1*

Preparation and Clean-up *page 2-2-1*

Survey *page 2-3-1*

Laboratory, Rack Room, and Wash Room *page 2-4-1*

Regulatory Treatment and Certification *page 3-1-1*

Control *page 4-1-1*

## Appendix A

Forms and Worksheets *page A-1-1*

## Appendix B

Emergency Aid and Safety *page B-1-1*

## Appendix C

Maintenance of the Manual *page C-1-1*

## Glossary

*page D-1-1*

## Index

*page Index-1*



# List of Figures

- FIGURE 2-1-1 Map of Golden Nematode Quarantine Areas in the State of New York *page 2-1-3*
- FIGURE 2-1-2 Mature Golden Nematode Cysts (right cyst showing larvae movement) *page 2-1-6*
- FIGURE 2-1-3 Mature Female Cysts on Potato Plant Roots *page 2-1-6*
- FIGURE 2-1-4 Field of Potato Plants Showing First Visible Signs of Golden Nematode Infestation *page 2-1-7*
- FIGURE 2-2-1 Summary of the Survey Process *page 2-2-3*
- FIGURE 2-2-2 Survey Supplies *page 2-2-15*
- FIGURE 2-2-3 Survey Supplies in Van *page 2-2-15*
- FIGURE 2-2-4 Example of Survey Sample Bag Labeling *page 2-2-17*
- FIGURE 2-2-5 Pressure Washing Equipment with Single Orifice Nozzle *page 2-2-19*
- FIGURE 2-2-6 Disinfecting Manual Survey Equipment *page 2-2-20*
- FIGURE 2-2-7 Pressure Washing Equipment and Supplies *page 2-2-22*
- FIGURE 2-2-8 Cleaning Equipment with a Single Orifice Nozzle *page 2-2-22*
- FIGURE 2-2-9 Sloped, Gravel Site for Cleaning Equipment *page 2-2-23*
- FIGURE 2-2-11 Sloped, Concrete and Gravel Pad for Equipment Cleaning *page 2-2-24*
- FIGURE 2-2-10 Pressure Cleaning Farm Equipment *page 2-2-24*
- FIGURE 2-3-1 Manual Soil Sampling *page 2-3-6*
- FIGURE 2-3-2 8 x 8 Sampling Method *page 2-3-7*
- FIGURE 2-3-3 Three-Wheeled Mechanical Sampler *page 2-3-9*
- FIGURE 2-3-4 Selection Guide for Collecting Soil Samples by Machine *page 2-3-10*
- FIGURE 2-3-5 Single Soil Samples and Supplies Loaded in Van for Transport to the Work Unit Laboratory *page 2-3-17*
- FIGURE 2-4-1 Rack Number and Shelf Number Label *page 2-4-2*
- FIGURE 2-4-2 Soil Samples Drying in the Rack Room *page 2-4-5*
- FIGURE 2-4-3 Sample Bag Beaker Number (50), Collection Number (DEM-03), Field Name (38-B-40), Bag Number (24), and Collection Date (1/11) *page 2-4-7*
- FIGURE 2-4-4 Beaker Numbers to Sample Bags *page 2-4-8*
- FIGURE 2-4-5 Fenwick Can Washers (Soil Sample Washing Machine) in Sample Wash Room *page 2-4-9*
- FIGURE 2-4-6 Opening Sample Bags *page 2-4-12*
- FIGURE 2-4-7 Beaker Number Confirmation *page 2-4-13*

FIGURE 2-4-8	Tank and Dumping Sample <i>page 2-4-14</i>
FIGURE 2-4-9	Sieves and Skimming Debris <i>page 2-4-15</i>
FIGURE 2-4-10	Sample Beakers on Laboratory Counter <i>page 2-4-17</i>
FIGURE 2-4-11	Sample Reading Station <i>page 2-4-20</i>
FIGURE 2-4-12	Golden Nematode Station <i>page 2-4-21</i>
FIGURE 2-4-13	Golden Nematode Laboratory Sample Processing Daily Sheet <i>page 2-4-22</i>
FIGURE 2-4-14	Pouring Sampling Material from Beaker into Muffin Tin Sieve <i>page 2-4-23</i>
FIGURE 2-4-15	Flotsam Examination for Cysts <i>page 2-4-24</i>
FIGURE 2-4-16	Golden Nematode Cysts <i>page 2-4-25</i>
FIGURE 2-4-17	Golden Nematode Cyst Placement in Vial <i>page 2-4-26</i>
FIGURE 2-4-18	Example of Vial Label <i>page 2-4-27</i>
FIGURE 3-1-1	Methyl Bromide Fumigation at Normal Atmospheric Pressure (NAP) <i>page 3-1-19</i>
FIGURE 3-1-2	Temperature for Dry Heat Fumigation of Soil Samples <i>page 3-1-23</i>
FIGURE 3-1-3	Temperature and Exposure for Steam Fumigation of Bench and Potting Soil <i>page 3-1-24</i>
FIGURE 3-1-4	Dosage, Temperature, and Reading for Methyl Bromide Fumigation at Normal Atmospheric Pressure (NAP), Chamber or Tarpaulin <i>page 3-1-26</i>
FIGURE 4-1-1	List of Golden Nematode Resistant Potato Varieties <i>page 4-1-3</i>
FIGURE A-1-1	Example of Golden Nematode Laboratory Sample Processing Daily <i>page A-1-3</i>
FIGURE A-1-2	Example of Golden Nematode Farm Survey Questionnaire (blank) <i>page A-1-7</i>
FIGURE A-1-3	Example of Golden Nematode Survey Data Worksheet <i>page A-1-9</i>
FIGURE A-1-4	Example of Golden Nematode Survey Data Worksheet <i>page A-1-10</i>
FIGURE A-1-5	Example of Sample Storage Sheet <i>page A-1-13</i>
FIGURE A-1-6	Example of Weekly Summary Record <i>page A-1-15</i>
FIGURE A-1-7	Example of PPQ Form 312, Golden Nematode Survey <i>page A-1-17</i>
FIGURE A-1-8	Example of GIS Image of Survey Site (ArcView) <i>page A-1-18</i>
FIGURE A-1-9	Hand-Drawn Diagram of Survey with Locations of Samples Collection <i>page A-1-19</i>
FIGURE A-1-10	Example of Hand-Drawn Nursery Survey Map <i>page A-1-20</i>
FIGURE A-1-11	Example of PPQ Form 333, Cyst Nematode Field Survey Log <i>page A-1-24</i>

- 
- FIGURE A-1-12 Example of PPQ Form 391, Specimens for Determination *page A-1-26*
- FIGURE A-1-13 Example of PPQ Form 519, Compliance Agreement (page 1 of 4) *page A-1-29*
- FIGURE A-1-14 Example of Attachment to Compliance Agreement for Potato Growers (page 2 of 4) *page A-1-30*
- FIGURE A-1-15 Example of Attachment to Compliance Agreement (page 3 of 4) *page A-1-31*
- FIGURE A-1-16 Example of Attachment to Compliance Agreement (page 4 of 4) *page A-1-32*
- FIGURE A-1-17 Example of PPQ Form 530, Limited Permit (blank) *page A-1-35*
- FIGURE A-1-18 Example of PPQ Form 540, Certificate of Federal/State Domestic Quarantines *page A-1-37*
- FIGURE B-1-1 Fumigant Route of Entry, Monitoring Device, and Exposure Source *page B-1-3*
- FIGURE B-1-2 First Aid Rescue Breathing *page B-1-5*
- FIGURE B-1-3 Signs and Symptoms of Fumigant Poisoning and Emergency Aid *page B-1-6*

## List of Figures

---

# List of Tables

- [TABLE 1-1-1](#) How to Report Problems with the Manual *page 1-1-4*
- [TABLE 1-1-2](#) How to Use Decision Tables *page 1-1-6*
- [TABLE 3-1-1](#) Determine if Certificate or Permit Is Required for Regulated Articles *page 3-1-7*
- [TABLE 3-1-2](#) Determine if Certificate or Permit Is Required for Used Containers and Used Equipment *page 3-1-17*
- [TABLE A-1-2](#) Instructions for Completing Golden Nematode Laboratory Sample Processing Daily *page A-1-5*
- [TABLE A-1-3](#) Instructions for Completing Golden Nematode Farm Survey Questionnaire *page A-1-8*
- [TABLE A-1-4](#) Instructions for Completing Golden Nematode Survey Data Worksheet *page A-1-11*
- [TABLE A-1-5](#) Instructions for Completing Sample Storage Sheet *page A-1-14*
- [TABLE A-1-6](#) instructions for the Weekly Summary Record *page A-1-16*
- [TABLE A-1-7](#) Instructions for Completing PPQ Form 312, Golden Nematode Survey *page A-1-22*
- [TABLE A-1-8](#) Instructions for Completing PPQ Form 333, Cyst Nematode Field Survey Log *page A-1-25*
- [TABLE A-1-9](#) Instructions for Completing PPQ Form 391, Specimens for Determination *page A-1-27*
- [TABLE A-1-10](#) Instructions for Completing PPQ Form 519, Compliance Agreement *page A-1-33*
- [TABLE A-1-11](#) Instructions for Completing PPQ Form 530, Limited Permit *page A-1-36*
- [TABLE A-1-12](#) Instructions for Completing PPQ Form 540, Certificate of Federal/State Domestic Quarantines *page A-1-38*

## List of Tables

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# 1

Golden Nematode  
Program Manual

# Introduction

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## Contents

Purpose	page 1-1-1
Scope	page 1-1-1
Users	page 1-1-3
Related Documents	page 1-1-3
Application	page 1-1-3
How to Use This Manual	page 1-1-3
How to Report Problems	page 1-1-4
Conventions	page 1-1-5

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## Purpose

The United States Department of Agriculture Animal, Plant Health Inspection Service (USDA-APHIS) takes regulatory action to prevent the spread of *Globodera rostochiensis* (golden nematode), which causes a dangerous disease of potatoes and certain other plants. In the United States, golden nematode currently infests certain counties or areas within counties in the State of New York **only**, and is neither widely prevalent nor distributed throughout the United States.

The *Golden Nematode Program Manual* will help prepare you for the following tasks:

- ◆ Performing detection and delimiting surveys for golden nematode in areas where the golden nematode is known or **not** known to occur
- ◆ Conducting treatments and certifying regulated articles for movement from regulated areas
- ◆ Determining the movement entry status of regulated and non-regulated articles
- ◆ Taking regulatory action when golden nematode is detected

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## Scope

### What the Manual Covers

The manual is divided into tabbed chapters:

- ◆ Introduction
- ◆ Procedures

- ◆ Control

The manual also contains appendixes, a glossary, and an index.

The **Introduction** provides basic information about the manual such as purpose, scope, and users. The introduction includes contact information for problems related to the manual, and also includes the a list of related documents, directions on how to use the manual, and a description of conventions (unfamiliar or unique symbols and highlighting) that appear throughout the manual.

The **Procedures** chapter contains the following information:

- ◆ History of the golden nematode
- ◆ Life cycle of the pest, host plants and infestations
- ◆ Detection and delimiting surveys
- ◆ Sample transport
- ◆ Sample washing
- ◆ Sample reading
- ◆ Preparing the suspect cysts for identification
- ◆ Cleaning and disinfection of supplies and equipment
- ◆ Directions for the certification of articles moving from golden nematode regulated areas
- ◆ Control methods used in the State of New York

The appendixes contain the following information:

- ◆ Examples and instructions for completing and issuing forms
- ◆ Safety procedures

The **Glossary** defines specialized words, abbreviations and acronyms, and other terms that are used which may be difficult or unfamiliar.

The **Index** contains topics and page numbers for quick reference.

### **What the Manual Does Not Cover**

The manual **does not** cover the following:

- ◆ Detailed information about the golden nematode
- ◆ Detailed pest identification procedures used by the golden nematode identification specialist

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## Users

This manual is written for use by PPQ Officers, Program Director, and Program Manager, and other Federal and State regulatory officers.

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## Related Documents

The authority for specific regulatory action is based on the Golden Nematode Quarantine 7CFR§301.85 and the Plant Protection Act. The State of New York Plant Regulatory Agency has enacted an interior parallel quarantine known as Part 127 of the Agriculture and Markets Law.

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## Application

This manual contains the policy, guidelines, and instructions that officers **must** follow as a basis for the treatment or other procedures to be used in authorizing the movement of regulated articles. This manual serves as a basis for explaining such procedures to persons interested in moving articles affected by quarantine regulations.

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## How to Use This Manual

Review the contents of this manual to get a feel for the scope of material covered. Glance through the section that you will be using, and familiarize yourself with the organization of the information. Use the table of contents which follows each tab to find the information you need.

**EXAMPLE**

To find information on the first signs of plant infestation, see [Plant Damage](#) on **page 2-1-6**.

If the table of contents is **not** specific enough, then turn to the index to find the topic and its page number.

## How to Report Problems

Use [Table 1-1-1](#) to report suggestions, problems, situations, and disagreements which directly affect the contents of the *Golden Nematode Manual*.

**TABLE 1-1-1 How to Report Problems with the Manual**

If you:	And the problem is:	Then:
Are <b>not</b> able to access the on-line manual	→	CONTACT the PPQ Manuals Unit John Patterson <john.l.patterson> @aphis.usda.gov or call 240-629-1934
Have a suggestion for improving the content of the manual	→	CONTACT the PPQ Manuals Unit, Deborah Briggs at deborah.j.briggs@aphis.usda.gov COMPLETE and MAIL the Comment Sheet (located at the back of the manual)
Have identified a problem with the content of the manual	<b>Not</b> urgent	COMPLETE and MAIL the Comment Sheet (located at the back of the manual)
	Urgent	CALL the Manuals Unit, either Deborah Briggs 240/629-1928
Have a situation that requires an immediate response regarding a procedure or regulatory action	→	CONTACT the Golden Nematode Program Director, Daniel Kepich, at daniel.j.kepich@aphis.usda.gov or call 607-566-2212
Disagree with a policy, procedure, or regulatory action identified in the manual	→	

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## Conventions

The conventions used in this manual are established by custom and are widely recognized and accepted.

### Advisories

Advisories are used throughout the manual to bring important information to your attention. Please carefully review each advisory. The definitions coincide with American National Standards Institute (ANSI), and are in the format shown below.



DANGER indicates that people could **easily** be hurt or killed.



WARNING indicates that people could **possibly** be hurt or killed.



CAUTION indicates that people could possibly be endangered or slightly hurt.



NOTICE indicates a possibly dangerous situation where goods might be damaged.



IMPORTANT indicates helpful information.

### Boldface

Boldfaced type is used to highlight negative or important words throughout this manual. These words are: **always, cannot do not, does not, except, must, never, no, not, only, other than.**

### Bullets

Bulleted lists indicate that there is **no** order to the information listed.

### Chapters

The manual has chapters divided into chapter sections. Every chapter and chapter section has a table of contents at the beginning that lists the heading titles within.

## Control Data

Information placed at the top and bottom of each page helps users keep track of where they are in the manual and updates to the manual. At the top of each page is the chapter and first-level heading. At the bottom of each page is the month, year, manual transmittal number, title, page number, and unit responsible for content.

## Decision Tables

Decision tables are used throughout the manual. The first and middle columns in each table represent conditions, and the last column represents the action to take after all conditions listed for that row are considered. Begin with the column headings and move left-to-right, then continue one row at a time.

**TABLE 1-1-2 How to Use Decision Tables**

<b>If you:</b>	<b>And:</b>	<b>Then:</b>
Read this column first	Continue in this row	TAKE the action listed in this cell
Read this column	Continue in this row	TAKE the action listed in this cell

## Examples

Examples are used to clarify a point by applying a real-world situation.

### EXAMPLE

Examples are graphically placed boxes within the text as a means of visually separating from other information on the page. Examples **always** appear in a box like this.

## Footnotes

Footnotes comment on or cite a reference to text and are referenced by number. Two types of footnotes are used in this manual: general text footnotes and table or figure footnotes.

General text footnotes are located at the bottom of the page and are consecutively numbered throughout the manual.

Table and Figure footnotes are located at the bottom of the associated single-page table or figure when space allows. However, for multi-page tables or tables that cover the length of a page, footnote numbers and footnote text **cannot** be listed on the same page. To locate footnote text, be sure to check the last page and the page following the end of the associated table or figure.

## Heading Levels

Within each chapter there are three heading levels. The first heading is indicated by a horizontal line followed by the title which continues across both the left and right columns. The second heading is

subordinate to the first heading, is in the right-hand column with the text beginning below it. The third heading is subordinate to the second heading, and located in the left-hand margin.

### Highlighting and Hypertext Links

When tables, figures, or other headings are cross-referenced in the body of the manual, they are emphasized in boldface and highlighted. Headings and titles are also italicized. These appear in blue hypertext in the on-line manual.

#### EXAMPLE

See [Figure 2-1-1 on page 2-1-3](#).

### Indentions

Entry requirements which are summarized from CFRs, permits, or policies are indented on the page.

### Italicized Brackets

When completing or reviewing certain certificates and forms, information that is to be entered, listed, or filled in is italicized and enclosed in brackets.

### Numbered Lists

Numbered lists are used to indicate the specific order in which the information listed is to be followed.

### Numbering Scheme

A two-level numbering scheme is used in this manual for pages, tables, and figures. The first number represents the chapter. The second number represents the page, table, or figure. This numbering scheme allows for easier updating and adding and removing pages without having to reprint an entire chapter. Dashes are used in page numbering to differentiate page numbers from decimal points.



# 2

Golden Nematode  
Program Manual

## Procedures

### *Introduction*

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#### Contents

Introduction	page 2-1-1
Golden Nematode Program	page 2-1-1
Cooperation With Other Agencies	page 2-1-2
Map of Quarantine Areas	page 2-1-3
General Pest Information	page 2-1-4
Distribution	page 2-1-4
Hosts	page 2-1-5
Life History	page 2-1-5
Golden Nematode Female Larvae	page 2-1-5
Adult Golden Nematode Males	page 2-1-5
Golden Nematode Cysts	page 2-1-5
Plant Damage	page 2-1-6

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#### Introduction

The *Introduction* section of the *Procedures* chapter provides information about the Golden Nematode Program, a map of areas in New York State that are currently regulated for golden nematode, and general information about the history of the pest, the life cycle, pest hosts, and plant and crop damage.

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#### Golden Nematode Program

The goal of the USDA-APHIS-PPQ Golden Nematode Program is to maintain a risk-based management system to prevent the spread of golden nematode (*Globodera rostochiensis*) and new infestations in potatoes, and to facilitate international and interstate agricultural shipments.

The Golden Nematode Program includes the following components:

- ◆ Certification of regulated articles for movement from regulated areas
- ◆ Decision-making on the movement of regulated and non-regulated articles into regulated areas
- ◆ Survey of regulated land
- ◆ Survey of fields planted with susceptible crop varieties
- ◆ Soil sample processing for golden nematode
- ◆ Control of golden nematode infestations and regulated land

- ◆ Treatment of equipment and supplies that enter, are used on, or exit regulated land

### **Cooperation With Other Agencies**

Sometimes the regulations of different Federal, State, and local agencies govern the same pests. As a result, USDA-APHIS-PPQ cooperates with the following Federal, State, and local agencies regarding golden nematode research, survey, and control.

#### **Federal**

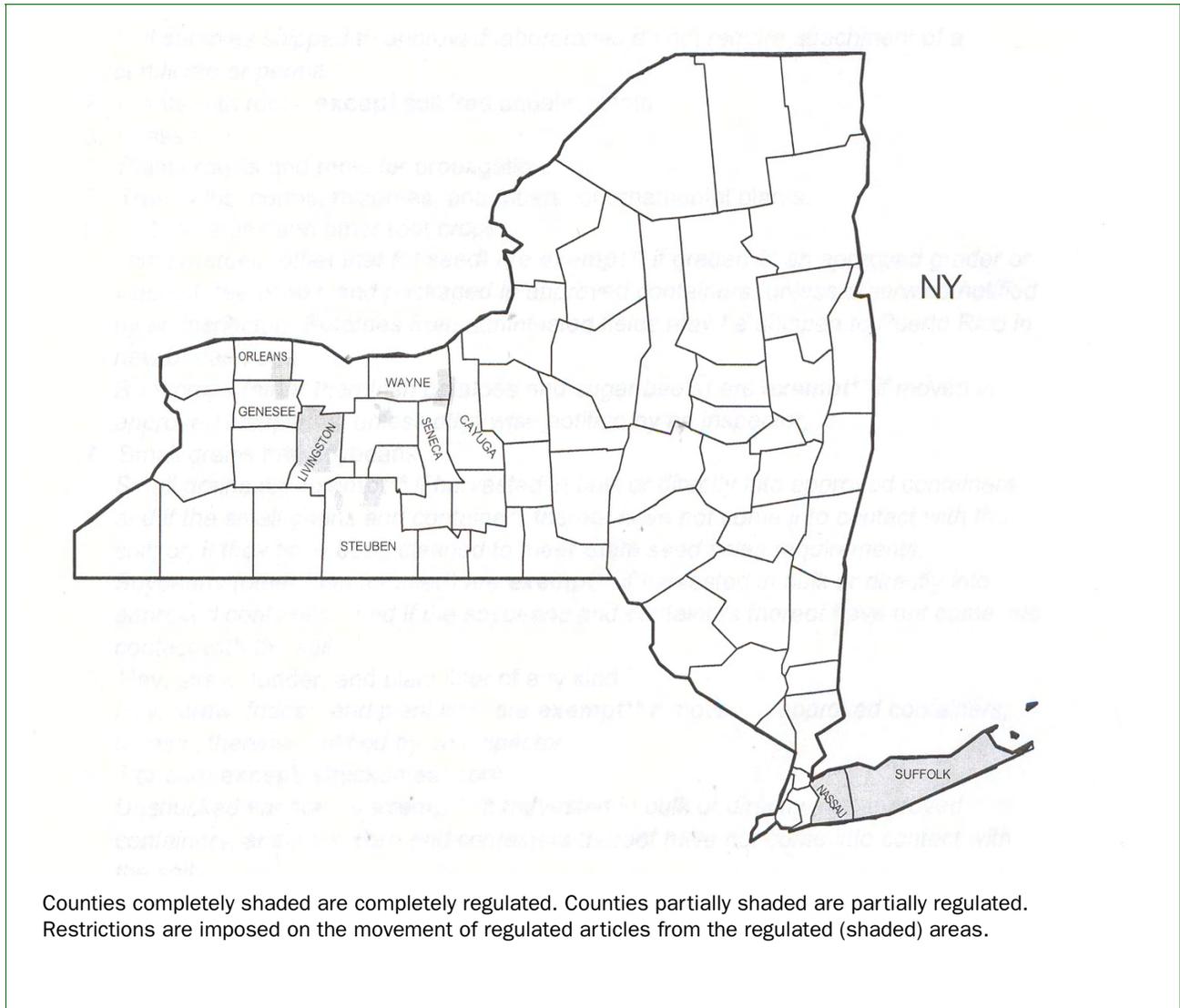
The USDA-Agricultural Research Service (ARS) provides assistance with the Golden Nematode Program.

#### **State and Local**

The New York Department of Agriculture and Markets shares regulatory responsibility for the Golden Nematode Program with APHIS. APHIS supports the development of golden nematode-resistant potato varieties at Cornell University through cooperative agreement. The New York Certified Seed Potato Improvement Cooperative provides research. The Extension Service of the Cooperative State Research Service provides assistance.

## Map of Quarantine Areas

The map in [Figure 2-1-1](#) shows the areas in New York State that are regulated for golden nematode as of January 26, 2004. For the most recent golden nematode quarantine map, go to the Web site <http://www.aphis.usda.gov/ppq/maps/>.



**FIGURE 2-1-1** Map of Golden Nematode Quarantine Areas in the State of New York

## General Pest Information

Of the many plant pests of foreign origin that have become established in the United States, *Globodera rostochiensis* (golden nematode) is potentially more dangerous than any of the other insects and diseases affecting the potato industry. Damaging populations of golden nematode develop when susceptible crops are planted in monoculture or rotation.

Potatoes and tomatoes are the principal crops of importance that are attacked by the golden nematode. Once golden nematode is established, the growing of potatoes and tomatoes **must** be done **only** by planting nematode-resistant varieties or in rotation with other crops for long periods. Continuous planting of non-resistant varieties of potatoes is impractical in golden nematode infested soil, due to decreases crop yield caused by increases in the nematode population.

### Strains

The primary golden nematode strain that infests land in New York is Ro1.

A second race of golden nematode, Ro2, has recently been detected in New York. Data collected indicates Ro2 develops on land infested with Ro1 where the same Ro1 potato-resistant variety.

### Distribution

In the United States, golden nematode Race 1 (Ro1) was discovered in 1941 on Long Island, New York. Infestations have since been confirmed in the New York counties of Cayuga, Genesee, Livingston, Nassau, Orleans, Seneca, Steuben, Suffolk, and Wayne.

New Castle County, Delaware previously infested, but has since been eradicated of golden nematode. Delaware was removed from quarantine in 1970.

Today the golden nematode is a major pest of potatoes in Europe. In England alone, nearly 75 percent of potato production land has severe crop restrictions due to golden nematode infestation. In addition to the Europe and the U.S., golden nematode has been also been found in parts of South America and Asia.

The first recorded Golden nematode infestation was in Germany in 1881. At the time, the golden nematode was considered to be a strain of *Heteroda schachtii*. By 1913, this nematode was discovered in Scotland. Finally in 1923, the golden nematode was described as a completely different species, and **not** a strain of *Heteroda schachtii*.

### Hosts

Although potatoes and tomatoes are the primary crops established to be golden nematode hosts, the golden nematode also reproduces on the roots of eggplants and on some wild solanaceous weeds.

Damaging populations of the nematode develop in infested fields when susceptible crops are planted in a monoculture or rotation. The pest will develop when fields of crops are planted with potatoes following potatoes, tomatoes following tomatoes, and potatoes following tomatoes, or tomatoes following tomatoes.

### Life History

Golden nematode eggs and larvae within cysts produced during previous infestations over-winter in the soil. When soil temperatures become favorable during spring and summer, the larvae begin to emerge from eggs in direct response to chemical exudates of host plant roots. As the larvae leave the cysts, they enter the soil, penetrate the host plant roots behind the root tip, and then migrate to a position near the host's vascular system where feeding begins. Larvae continue to emerge from cysts throughout the growing season, and golden nematodes in various stages of development can be found in and on host roots. Normally **only** one golden nematode generation is produced per year in the New York temperature zone.

### Golden Nematode Female Larvae

As the developing female larvae enlarge and break through the surface of the roots of the host plant, they remain attached to the host plant roots by their necks. Females pass through pearly-white and yellow color phases and retain eggs within their bodies.

The female nematode remains attached to host-plant roots while being fertilized by the adult male nematode. At death, females become the brown cysts which are easily detached from host-plant roots and remain in the soil after harvest of the host-plant crop.

### Adult Golden Nematode Males

Adult males are worm-like. They separate from the roots of the host-plant to search, find, and fertilize the female larvae.

### Golden Nematode Cysts

A golden nematode cyst is the dead body of a female nematode. The cyst is a spheroid, brown, thick-walled structure, and small enough that several cysts will fit on a pinhead. The cyst gives considerable

protection for the eggs and larvae within. Each cyst can contain up to 500 eggs and larvae. The eggs inside these cysts can remain viable at least 20 years.



**FIGURE 2-1-2 Mature Golden Nematode Cysts (right cyst showing larvae movement)**

### Plant Damage

Golden nematodes bore into the roots of host plants and feed on the plants' juices. This feeding **does not** cause immediate damage the above-ground part of the infested plant, and consequently, infestation often goes undetected for years.



**FIGURE 2-1-3 Mature Female Cysts on Potato Plant Roots**

The first sign of infestation is usually poor plant growth in one or more areas of the potato, tomato, or eggplant field. Signs of infestation include wilting, stunted growth, poor root development, and early death of the plant. As nematode populations increase, poor plant growth areas enlarge and newly-damaged areas appear on plants in the field. Eventually, the entire field shows poor plant growth.



Photo courtesy of Daniel Kepich

**FIGURE 2-1-4 Field of Potato Plants Showing First Visible Signs of Golden Nematode Infestation**



# 2

Golden Nematode  
Program Manual

## Procedures

### *Preparation and Clean-up*

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#### Contents

Introduction	<b>page 2-2-1</b>
Soil Survey	<b>page 2-2-2</b>
Survey Preparation	<b>page 2-2-3</b>
Step 1: Confirm the Reason for the Survey Is Valid	<b>page 2-2-3</b>
Step 2: Determine the Type of Survey to Conduct	<b>page 2-2-3</b>
Step 3: Prepare for the Survey	<b>page 2-2-5</b>
PPQ Plant Health Safeguarding Specialist (PHSS)	<b>page 2-2-5</b>
PPQ Technician Responsibilities	<b>page 2-2-7</b>
Crew Leader Responsibilities	<b>page 2-2-7</b>
Step 4: Assemble the Survey Crew	<b>page 2-2-8</b>
Crew Leader Responsibilities	<b>page 2-2-7</b>
Step 5: Conduct the Survey	<b>page 2-2-10</b>
	<b>page 2-2-11</b>
	<b>page 2-2-11</b>
Manual Survey Field Crew	<b>page 2-2-12</b>
Mechanical Survey Tractor Operator	<b>page 2-2-12</b>
Survey Materials and Equipment	<b>page 2-2-14</b>
Materials	<b>page 2-2-14</b>
Survey Equipment	<b>page 2-2-16</b>
Sample Bag Labeling	<b>page 2-2-16</b>
Sanitation	<b>page 2-2-18</b>
Survey Equipment	<b>page 2-2-16</b>
Cleaning Vehicles	<b>page 2-2-18</b>
Special Instructions for Cleaning of Clothing, Equipment and Supplies During Survey of Commercial Potato Fields	<b>page 2-2-18</b>
Cleaning and Disinfection for Survey of Seed Potato Fields	<b>page 2-2-19</b>
Cleaning and Disinfection for Confirmatory and Other Surveys	<b>page 2-2-20</b>
Cleaning and Disinfecting Materials and Supplies	<b>page 2-2-21</b>
Crew Leader	<b>page 2-2-21</b>
Cleaning and Disinfecting Equipment	<b>page 2-2-21</b>
Steam Heat Treatment	<b>page 2-2-23</b>

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#### Introduction

The *Preparation and Disinfection* section of the *Procedures* chapter provides information that will help personnel get ready to conduct a survey, procedures to follow during a survey, instructions for proper cleaning and disinfecting of supplies, equipment, and vehicles after the survey is finished.

## Soil Survey

Soil survey for golden nematode is conducted to determine whether or **not** the soil is infested. In the State of New York, survey for golden nematode is currently conducted every place that grows potatoes.

### Exposed Land

Exposed land is land that meets any of the following criteria:

- ◆ Land that was infested and has been fumigated, resurveyed, and released from quarantine regulation prior to 1972<sup>1</sup>
- ◆ Land operated by a farmer who has farmed infested land
- ◆ Land farmed with equipment used in a field with a history of infestation
- ◆ Land bordering a field with a history of infestation
- ◆ Land that receives direct drainage from a field with a history of infestation
- ◆ Land exposed as a result of a regulatory violation

### Non-exposed Land

Non-exposed land and fields are those that have **never** had golden nematode infestation. Non-exposed fields are sampled and tested for golden nematode when the fields are planted with susceptible variety of potatoes, tomatoes, or eggplant in a county or area that is regulated for golden nematode.

Where surveying all potato or tomato fields is **not** possible, selection of fields to survey should be based on crop history. Select fields which have been planted to potatoes and/or tomatoes consecutively for the longest period of time.

There is little likelihood of finding nematode in fields planted to a host crop for less than three (3) consecutive years. If crop rotation is practiced on a field, then a longer period of time (more than 3 consecutive years) is required for the nematode to build up to a detectable level.

### Soil Conditions

If the soil is too wet, then delay survey until ground is drier.

<b>EXAMPLE</b>	If you can take a clump or handful of soil and squeeze the soil into a ball, then the soil is too wet to sample.
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1 All land found infested from 1972 to the present is regulated by New York State Part 127, Golden Nematode Quarantine. Under New York State Law, this land is officially called regulated land.

## Survey Preparation

Before you begin a survey, follow the procedures listed in [Figure 2-2-1](#) below.

1. Confirm the reason for the survey is valid.
2. Determine the type of survey to conduct.
3. Assemble the survey crew.
4. Prepare materials, equipment, for the survey.
5. Conduct the survey.
6. Disinfect survey materials and equipment.
7. Transport samples to laboratory.

**FIGURE 2-2-1 Summary of the Survey Process**

### Step 1: Confirm the Reason for the Survey Is Valid

The process to conduct a survey for golden nematode when any of the following conditions occur:

- ◆ Farmer request for export certification for freedom from golden nematode (voluntary)
- ◆ Exposure of land to possible golden nematode infestation (mandatory)
- ◆ Potato varieties susceptible to golden nematode are planted and grown three (3) years or more (consecutively or in rotation) (mandatory)
- ◆ Regulatory violation (someone moves equipment from an area that has golden nematode to an area that does not have golden nematode infestation) (mandatory)
- ◆ Seed potato certification (grower needs to certify potatoes for export certification or interstate movement) (mandatory)

Confirm the reason to conduct the survey falls under a category listed above. Continue to [Step 2: Determine the Type of Survey to Conduct](#).

### Step 2: Determine the Type of Survey to Conduct

Field soil sampling is used for detection and delimiting purposes. The pattern and number of samples to be taken will vary depending upon the likelihood of infestation, acreage involved, personnel available, and other factors.

The first-line supervisor or Program Manager will decide which type of survey to conduct. Determine which survey to conduct, based on the results of the initial survey or confirmation survey.

1. (Prior to beginning the survey work, contact all growers involved in the survey to inquire and determine if the growers have encountered any trouble spots in potato or tomato fields. (See [Plant Damage on page 2-1-6.](#))
2. Visit the site.
3. If soil is too wet, then **do not** survey yet.
4. Obtain a GIS map of the site.

The survey types available are:

- ◆ Confirmation survey
- ◆ Grader survey
- ◆ Manual survey (systematic manual soil sampling)
  - ❖ 8-by-8 block method
  - ❖ Simplified 8-by-8 block method
  - ❖ Modified 8-by-8 (4-by-8) block method
- ◆ Mechanical survey (mechanical wheel soil sampling)
- ◆ Nursery survey
- ◆ Peripheral survey
- ◆ Post crop survey
- ◆ Post resistant variety treatment survey
- ◆ Requested field survey
- ◆ Selected area soil sampling
- ◆ Symptom survey
- ◆ Survey to release land from exposed status
- ◆ Survey of seed potato production areas
- ◆ Survey outside regulated areas

See [Survey on page 2-3-1](#) for more information on each survey listed above.

### Step 3: Prepare for the Survey

Staff will follow the procedures listed below to prepare for the survey.

#### PPQ Plant Health Safeguarding Specialist (PHSS)

The Plant Health Safeguarding Specialist (PHSS) responsibilities for the assigned area include the following:

1. Interview the grower to obtain information and complete the Golden Nematode Farmer Survey Questionnaire (Pre-Survey Grower Questionnaire) prior to the survey. See **Golden Nematode Farm Survey Questionnaire** on page A-1-7.
2. Use the data from the Golden Nematode Farmer Survey Questionnaire to complete the Golden Nematode Survey Data Sheet.
3. Set up the survey.
4. Complete as much information as possible on PPQ Form 312, in advance. See **PPQ Form 312, Golden Nematode Survey** on page A-1-17.
5. Obtain an aerial map of the field to be surveyed, as follows:
  - A. If available, use GIS software to obtain the map (i.e., ArcView, Google Maps, etc.). GIS maps provide the highest quality and greatest detail. (See <http://www.aphis.usda.gov/gis/homepage.htm> or <http://www.Google.maps.satellite>.)
  - B. If a GIS map is **not** available, then use an aerial photographic map, topographic map or a good hand-drawn map showing clear details (road names, landmarks, etc).
6. Place the map on the back of *PPQ Form 312, Golden Nematode Survey*, and indicate the NW latitude and longitude on the map.
7. Coordinate all survey and regulatory responsibilities in the assigned area of coverage.
8. Oversee PPQ Technicians and temporary personnel hired to conduct the survey sampling.
9. Oversee the survey.
10. Check the weather status and determine if conditions are acceptable to conduct survey.
  - A. If raining, then **do not** conduct the survey.
  - B. If soil is too wet, then **do not** conduct the survey.
  - C. If the survey will **not** be conducted due to weather, then notify crew leader and survey crew.
11. Review the completed PPQ Form 312, at end of each day.

- 12.** File the completed PPQ Form 312 in the Record of Infestation folder, which is filed by county. The form will remain in the folder while the sample are in storage and drying.

### PPQ Technician Responsibilities

The PPQ Technician's responsibilities include the following:

1. Obtain maps from PHSS or prepare maps for next day's sampling.
2. Assemble the survey crew.
3. Perform crew leader duties.
4. Assist the PPQ Plant Health Safeguarding Specialist (PHSS).

### Crew Leader Responsibilities

The crew leader (PPQ-PHSS, PPQ Technician, or a temporary personnel) will conduct the following before departing the USDA facility each day:

1. If raining, then check with the PPQ-PHSS to determine whether or **not** survey will be conducted.
2. Gather supplies (steno notebook and pencil to sketch field). If supplies have been gathered the at the end of the previous workday, then verify everything is in the survey vehicle, including fresh water.
3. Give each crew member a copy of the field *Golden Nematode Survey Manual*, which includes the following information:
  - ❖ Crew guidelines
  - ❖ Expense log
  - ❖ Golden Nematode Quarantines Map and list of regulated articles that require a certificate or permit year-round
  - ❖ Fact sheet: Golden Nematode A Pest of Importance
  - ❖ Flyer: Reduce Your Risk of Tractor Overturn
  - ❖ Flyer: What You Need to Know About Preventing Plant Poisoning
  - ❖ Important phone numbers list
  - ❖ Local hospital's Poison and Drug Information Center phone number (taped to the inside binder)
  - ❖ Paper, blank (5)
  - ❖ PPQ Form 312, Golden Nematode Survey (3)
  - ❖ PPQ Form 333, Cyst Nematode Field Survey Log (3)
  - ❖ Reminders
  - ❖ Safety procedures from the Avoca Work Unit Safety & Health Office
  - ❖ Tractor operator guidelines
4. Determine how many fields to survey.

5. Distribute the field maps and identify the field to survey.
6. Review the survey maps.
7. Supervise initial stocking of supplies in transport vehicle.
8. Distribute boots (outfit) for each crew member.
9. Distribute bags and markers to number the samples on the way to the site.

#### **Step 4: Assemble the Survey Crew**

The PPQ Technician will assemble the survey crew. A typical field crew for a manual survey is 3 people, but the number of crew members may vary from 1 to 7 people, depending on the size and number of fields to sample, and the survey type.

#### **Survey Crew Responsibilities**

The survey crew will do as follows:

1. In the event of bad weather, the GN Survey Crew should call the officer-in-charge of their area before coming to work. If the officer-in-charge **cannot** be reached, then call the Avoca office.
2. A vehicle which can be washed, cleaned, and disinfected (such as a work-type van without carpeting or a covered-bed pickup truck is preferred. If a vehicle with carpeting must be used, then place plastic sheathing over the seat back and on the carpeted area where samples will be loaded.
3. Prepare the truck or van for the trip to sample site.
  - A. Gather supplies.
  - B. Confirm tools, bags, and equipment are clean.
  - C. Arrange supplies in the rear of a covered-bed pickup truck or van in an organized manner so that items can be easily removed from the vehicle.
  - D. Leave space toward the rear of the vehicle so that paper sample bags can be stacked side to side.
4. Review the survey maps.
5. Help label bags on the way to the survey site (see **Sample Bag Labeling on page 2-2-16**).
6. Help clean and restock the survey van or truck with supplies for next day's work.

## Tractor Operators

Tractor operators will do as follows:

1. Daily, know where you are going to sample and post the sampling location on the schedule board.
2. Daily, make sure you have all equipment and supplies for daily operation. Ensure that tools are not misplaced during the workday.
  - A. Check the tool box assigned to your tractor each morning before departing for the sampling location.
  - B. Check the tool box assigned to your tractor at the end of the day with your supervisor.
  - C. Remove the tool box and tools from your tractor and store in the tractor shed as directed. **Do not** leave tools on the tractor overnight.
  - D. When samples are taken to the rack room, make sure the storage rack sheets are filled out.
3. Follow tractor operation and maintenance guidelines. As a tractor operator, you are responsible for the safe operation and routine maintenance of the equipment assigned to you.
  - A. First day of each week:
    - i. Check the oil level; if needed, add oil.
    - ii. Check the transmission fluid; if needed, add fluid.
    - iii. If applicable, check the battery fluid level.
  - B. Daily:
    - i. Check the oil level; fill as needed.
    - ii. Add gas while the tank is cold and the engine is off.
    - iii. Check the trailer tires, tie-downs, lights, and secure safety chain to be sure they are in working order.
4. Operate the tractor safely, as follows:
  - A. Keep the tractor at a speed low enough to prevent bouncing.
  - B. Do not attempt sharp turns at high speed.
  - C. If driving the tractor on a highway, then lock the brakes together.
  - D. Use flashers and lights on roads and highways.
  - E. Use farm roads to enter and leave fields.
  - F. If operating the tractor in the field, then unlock the brakes.
  - G. At field ends, raise the equipment before turning.
  - H. Make end turns slow, brake-assisted, but **not** locked.

5. Boots **must** be worn in the fields and washed at the same time as the tractor.
6. Carry PPQ Form 312, Cyst Nematode Survey, with you and fill out. Have the form completed when the collection is done. Give the completed form to your supervisor.
7. The tractor must be washed before moving between divided and/or different growers' fields. Open the tractor's battery box; clean the battery box and the extra punches.
8. Let the supervisor know when you need the tractor and equipment, and your boots cleaned off and washed so that you can move to a new field.
9. Clean and secure the tractor at the end of each work day.
10. No smoking is allowed in vehicles or Government buildings.
11. Obtain supervisors approval before having lunch in restaurants or dinners during the work day.

### **Step 5: Conduct the Survey**

In addition to the procedures listed under [Survey on page 2-3-1](#), follow the steps listed below at the survey site.

#### **Crew Leader**

At the sampling site, the crew leader will do as follows:

1. At the sampling site, park the transport vehicle and place 1 orange traffic cone in front of the vehicle and 1 traffic cone at the rear of the vehicle.
2. Put boots on before entering field, and make sure crew members have their boots do the same.
3. Use the GIS map on back of PPQ Form 312, Golden Nematode Surve. Use the reading NW (latitude, longitude) (Goggle/maps/field). at least 2 points use Shape file (satellite photos) Arc view.
4. Walk to the field and use your judgment to determine which end to start sampling.
5. If manual survey, carry mesh tote bag/backpacks containing the samples and trowels, and place the bags at the beginning and end of each row to collect filled sample bags inside.
  - A. If mechanical survey, the tractor operator will remove the bags from the bag holders, fold and staple each bag, and place the bags in the bag basket.
6. Complete PPQ Form 312.
7. Use a GIS photo of the field and sketch the location (from which each the sample is obtained. If a GIS photo is not available, then sketch in landmarks, fences, mailbox, and road names.
8. Conduct/assist with the sampling.

9. If conducting a manual survey, as each paper sample bag is filled, pick up the sample bags, fold the top down, staple, and place each bag inside the nearest mesh tote/backpack.
10. Make sure all crew members keep their boots on until sampling is finished.
11. Remove soil sample bags from mesh tote bags/backpacks and stack in consecutive order in the transport vehicle.

### Crew Leader

After the survey is finished, the crew leader will make sure the supplies are cleaned and disinfected as follows:

1. Set up 3 stations for disinfecting supplies; prepare a solution of one part 5.25 hypochlorite (bleach) to 10 parts of water.
2. Have each person remove their boots at the vehicle; and **do not** anyone to re-enter the field after boots have been removed.
3. Have each person use the stiff bristle brush to remove soil from books, trowels, and mesh tote bags/backpacks.
4. Clean and disinfect tractors and pickup tracks at the survey site. See [Cleaning Vehicles](#) on page 2-3-4.
5. If a van is used to transport samples, then stop at a carwash to wash the van on the way back to the lab from the survey site.

Back at the laboratory, after unloading the samples, do as follows:

1. Supervise the cleaning and disinfection of survey transport vehicle used during day.
2. Supervise or assist the restocking of supplies in the transport vehicle.
3. Make a copy of the PPQ Form 312 completed that day.
4. Give the completed PPQ Form 312 to the PHSS or PPQ Technician at the end of each day. (The PHSS will give the form to the PHSS to review and make sure all blocks are completed. The PHSS will give the form to the Laboratory Leader at end of each day.)
5. Get the next day's maps from the PPQ technician and distribute to crew members the sampling day morning.
6. Make sure the vehicle is clean and has been washed at carwash to remove soil from undercarriage.

### **Manual Survey Field Crew**

The field crew will do as follows:

1. Put your boots on before entering field.
2. If survey, walk to the field.
3. Start sampling where directed by crew leader.
4. Place filled sample bags in mesh tote/backpack.
5. Keep boots on and walk to vehicle.
6. Remove each sample bag from the mesh backpack/tote and staple closed.
7. Load soil samples from mesh tote bags/backpacks, stacking sample bags in consecutive order in the transport vehicle, If a van or other carpeted vehicle is used to transport vehicles, then make sure plastic is placed over the carpet before the sample bags are loaded into the vehicle.
8. Confirm boots, trowels and mesh bags have been thoroughly cleaned with bleach solution before putting trowels and boots in clean container and placing in transport vehicle.
9. Empty and rinse buckets, containers, and brushes, and clean and disinfect with bleach solution before placing back into the transport vehicle.
10. Empty wash water onto the same side of the road as the field surveyed so that the water will drain back into the survey area.
11. After survey sampling for the day, transport the sample bags to the laboratory.
12. Clean and disinfect vehicle,
13. Prepare the transport vehicle for the next day's use.

### **Mechanical Survey Tractor Operator**

The mechanical survey is completed by the tractor operator. The tractor operator will do as follows:

1. Review the survey map with the PPQ Technician and discuss conditions and where to park.
2. Operate the tractor in a safe manner.
3. Unload the tractor from the transport trailer.
4. Place prenumbered bags in bag holders on the mechanical sampling equipment.
5. Watch bags so that each is no more than a third to a half filled with the sample.
6. Stop the tractor, remove each bag, fold and staple, place filled bags in the bag basket, and safeguard the sample bags.
7. Place new bags in the sample bag collection holder.

8. Keep PPQ Form 312 and fill in.
9. Clean and disinfect the tractor and trailer.

---

## Survey Materials and Equipment

### Materials

Materials needed for the field survey are listed below:

- ◆ Boots, neoprene, (1 pair for each crew member and a few spare pair)
- ◆ Brushes, stiff bristle
- ◆ Clipboard
- ◆ First Aid kit
- ◆ Hypochlorite 5.25 percent (bleach)
- ◆ Maps
- ◆ Paper bags, heavy duty
- ◆ Permanent markers (red, green, and black)
- ◆ Plastic tubs (for disinfecting shoes)
- ◆ PPQ Form 312, Golden Nematode Survey
- ◆ Raincoats (for equipment cleaning or steam cleaning)
- ◆ Rubber gloves, lined (3-7 pair)
- ◆ Safety cones
- ◆ Sponges
- ◆ Stapler, heavy duty
- ◆ Traffic cones (2 for manual survey)
- ◆ Trowels (long-handled)
- ◆ Water container (filled with clean drinking water)

See the survey supplies in [Figure 2-2-2 on page 2-15](#) and.



Photo by Dan Kepich

**FIGURE 2-2-2 Survey Supplies**



Photo by Dan Kepich

**FIGURE 2-2-3 Survey Supplies in Van**

## Survey Equipment

The following equipment is needed:

- ◆ Low pressure pump
- ◆ Pick-up truck
- ◆ Tank of water
- ◆ Tractor (for mechanical survey)
  - ❖ 2-wheel sampling equipment (use on non-exposed field (no previous GN infestation))
  - ❖ 3-wheel sampling equipment (use on exposed field)
- ◆ Trailer (for mechanical survey to carry samples, weather permitting; pulled by the truck that carries pressure washing cleaning equipment)
- ◆ Truck (preferred) or van **without** carpet (for cleaning and disinfection)

## Sample Bag Labeling

Regardless of the type of sampling followed, all bags used **must** be properly labeled and accurate records prepared and maintained.

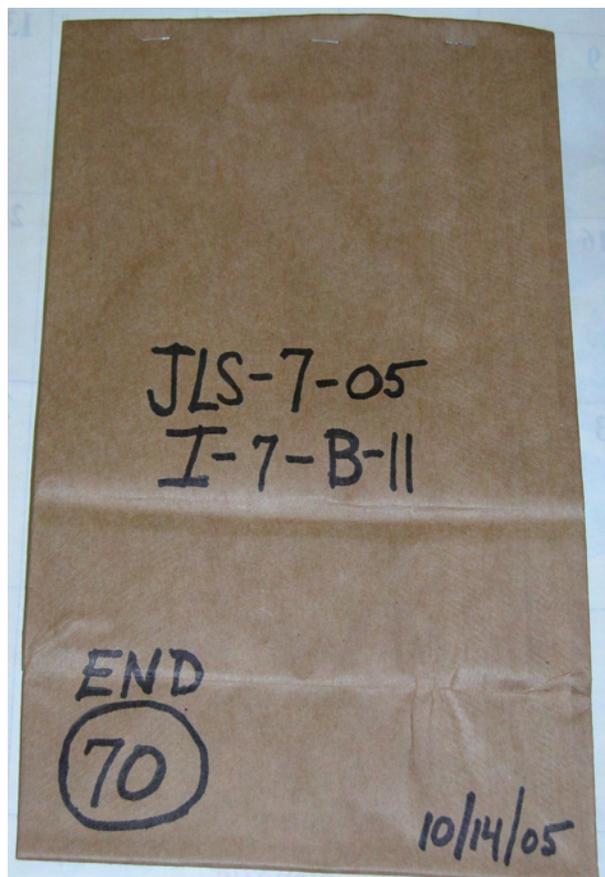
Label each bag with the following information starting just below the top fold of the bag:

1. Use the designated permanent marker color:
  - ❖ Red ink: Ro2 and post-treatment samples
  - ❖ Green ink: certified seed potato land samples
  - ❖ Black ink: all other samples
2. Record the crew leader's initials, consecutive collection number, and year on the first line in the center of the bag. The crew leader **must**:
  - ❖ Exercise caution so that numbers are **not** duplicated
  - ❖ Continue with consecutive numbering through the same calendar year (even when moving the sampling to another township, county, or State)
  - ❖ **Not** start new numbering until the beginning of the next calendar year

<b>EXAMPLE</b>	JJD-1-06 (for John J. Doe, collection 1, in 2006)
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3. Record the farmer's name and the map designation or field number on the second line.

4. Record the date (month, day, and year) in the lower right hand corner. A
5. Record the number of each the individual bag (each sample bag has a separate number) making up the collection in a given location in the lower left-hand corner and circle the number, beginning with 1 for the first sample bag in the collection, and numbering each sample bag consecutively. Write "END" above the last sample bag number in the collection.



**FIGURE 2-2-4 Example of Survey Sample Bag Labeling**

## Sanitation

Particular care **must** be taken when working in golden nematode infested fields. Each person and piece of equipment involved in program activities of the golden nematode quarantine is a potential carrier of this pest. Every reasonable precaution **must** be taken to prevent the spread of golden nematode by program personnel or equipment.

### Cleaning Vehicles

Vehicles (cars, tractors, trucks, etc.) should remain on hard-surface roads. When driving vehicles on field roads is necessary, then the vehicles **must** be thoroughly sanitized as follows before leaving the survey site:

1. Remove all soil from vehicles and any equipment used. Power washing equipment will be made available as standard equipment for removing soil from vehicles.
2. Clean the vehicles and equipment with steam or water under pressure using a single orifice nozzle.
3. If the survey is of seed potato production areas, then additional sanitary procedures are required.
4. Mechanical survey equipment which has been used on infested or exposed land must **never** be used seed potato production land.



**No** vehicles or mechanical soil sampling equipment can be used or enter seed potato production land. **No** equipment for golden nematode survey that has been used on infested or exposed land can be used on seed potato production land.

### Special Instructions for Cleaning of Clothing, Equipment and Supplies During Survey of Commercial Potato Fields

For survey of commercial potato fields, each cleaning **must** be completed as follows before leaving one field and continuing to the next field:

1. Provide each crew member with a stiff bristle brush.
2. Each crew member will use the brush to clean all soil from trousers, shoes, and equipment.
3. Thoroughly clean trowels to be used for sample collection. Trowels **must** be free of recesses or grooves where soil might become impacted.

4. Before leaving a property, clean mechanical sampling equipment with steam or water under pressure using a single orifice nozzle to remove all soil from the equipment.



Photo by Dan Kepich

**FIGURE 2-2-5 Pressure Washing Equipment with Single Orifice Nozzle**

### **Cleaning and Disinfection for Survey of Seed Potato Fields**

All surveys conducted on seed potato land will be accomplished by manual survey procedures.

Personnel sampling seed potato lands will be provided with lightweight neoprene boots to wear as follows.

1. Tuck trouser or pant legs inside these boots before entering the sampling field and keep tucked while samples are being collected.
2. Before entering each field and again when leaving each field, clean and disinfect all boots and equipment as follows:
  - A. Use a stiff bristle brush to remove all soil.

- B.** Scrub with a solution of 5.25 percent hypochlorite in 10 parts water.



Photo by Steve Kupper

**FIGURE 2-2-6 Disinfecting Manual Survey Equipment**

### **Cleaning and Disinfection for Confirmatory and Other Surveys**

Sanitary procedures outlined for seed potato fields will be followed while conducting confirmatory surveys and at any other time the inspector deems appropriate.

## Cleaning and Disinfecting Materials and Supplies

### Crew Leader

After the survey is finished, set up a cleaning site that is near the sampling site, and sloping downward. follows:

1. Set up 3 stations for disinfecting supplies; prepare a solution of one part 5.25 percent hypochlorite (bleach) to 10 parts water in tubs.
2. Remove your boots at vehicle; and **do not** re-enter the field after boots have been removed.
3. Use a stiff bristle brush to remove loose soil from boots, trowels, and mesh tote bags/backpacks.
4. Clean and disinfect boots, trowels, and mesh bags using bleach solution and brush.
5. Empty the used cleaning solution to drain back into the field.
6. If the vehicle's interior has any carpeting, then thoroughly vacuum the carpet to prevent soil contamination.
7. Place supplies back in vehicle.

## Cleaning and Disinfecting Equipment

A concrete pad, blacktop area, or driveway is the ideal location for cleaning equipment; otherwise, a gravel area or a grassy area right next to the field sampled can be used. The location of the cleaning site **must** be sloping downward so that the runoff will drain back into the field.

1. Move the equipment to the cleaning site.
2. Hook up the low pressure pump (small pump) to the tank of water, and use the low pressure pump to get most of the soil off first,
3. Use the high pressure washer to finish cleaning the equipment.

Equipment is cleaned under the following conditions:

- ◆ After a survey or after entering regulated land
- ◆ Compliance agreement terms
- ◆ Request from farmer or landowner to clean equipment
- ◆ Someone enters and plans to exit golden nematode regulated land or a field with equipment (must be cleaned before moving from field to field or existing an infested field and moving back onto the road)
- ◆ Well-drilling equipment (wells drilled on GN regulated land)
- ◆ Grower/farmer contacts USDA about sale of equipment

- ◆ Custom farming equipment is used on regulated land (specialized business where a specific service is performed from farm to farm - moves; such as fertilizer applicators, lime applicators)
- ◆ Drainage ditch digging equipment is used on regulated land (drainage ditches dug are dug on GN infested land)



**FIGURE 2-2-7 Pressure Washing Equipment and Supplies**



**FIGURE 2-2-8 Cleaning Equipment with a Single Orifice Nozzle**

### **Pressure Wash Treatment**

Pressure wash tractors, tractor equipment, farm implements, cultivators, pickup trucks. then clean hoses, boots, raincoats, and anything else that could potentially move golden nematode cysts from a regulated field to a non-regulated field.

### **Steam Heat Treatment**

See the PPQ Treatment Manual for equipment needed and description of steam heat treatment.



Photo by Dan Kepich

**FIGURE 2-2-9 Sloped, Gravel Site for Cleaning Equipment**



Photo by Dan Kepich

**FIGURE 2-2-10 Pressure Cleaning Farm Equipment**



Photo by Dan Kepich

**FIGURE 2-2-11 Sloped, Concrete and Gravel Pad for Equipment Cleaning**

# 2

Golden Nematode  
Program Manual

## Procedures

### Survey

#### Contents

Introduction	page 2-3-2
Surveys in New York State	page 2-3-2
Survey of Seed Potato Production Areas	page 2-3-3
Soil Surveys	page 2-3-3
Sanitation	page 2-3-4
Cleaning Vehicles	page 2-3-4
Cleaning and Disinfecting Survey Materials	page 2-3-5
Cleaning of Clothing, Equipment, and Supplies During Survey of Commercial Potato Fields	page 2-3-5
Cleaning and Disinfection for Survey of Seed Potato Fields	page 2-3-5
Cleaning and Disinfection for Confirmatory and Other Surveys	page 2-3-5
Systematic Manual Soil Sampling	page 2-3-6
8-by-8 (8 x 8) Block Method	page 2-3-7
Simplified 8-by-8 Block Sampling Method	page 2-3-8
Modified 8-by-8 (4-by-8) Block Sampling Method	page 2-3-8
Mechanical (Wheel) Soil Sampling	page 2-3-9
Selected Area Soil Sampling	page 2-3-10
Peripheral Survey	page 2-3-11
Grader Survey	page 2-3-11
Nursery Survey	page 2-3-11
If collecting from piles of potting soil, take a sample from each pile and mark the location of each sample's origin on the sample bag. Survey Times and Types	page 2-3-11
Post-crop Survey	page 2-3-12
Rested Field Survey	page 2-3-12
Confirmation Survey	page 2-3-12
Post Resistant-Variety Treatment Survey	page 2-3-13
Survey to Release Land from Exposed Status	page 2-3-14
Survey Outside Regulated Areas	page 2-3-14
Symptom Surveys	page 2-3-14
Surveys Outside of New York State	page 2-3-15
Sample Collection Bag Transport	page 2-3-16
Sample Transport Vehicles	page 2-3-16
If a program does not yet have a vehicle dedicated to seed potato soil sampling transport only, then another vehicle may be used provided the interior and exterior has been completely disinfected with steam heat treatment. Loading Sample Collection Bags	page 2-3-16

## Introduction

The *Survey* section of the *Procedures* chapter provides procedures for conducting systematic soil surveys, labeling samples, and disinfecting soil sampling equipment. Survey procedures apply to both GN Ro1 and GN Ro2.

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## Surveys in New York State

The general guidelines listed in the section apply to **all** survey regions in the State of New York and all surveys will be conducted in accordance with this manual.

Routine soil surveys will **not** be conducted on land with less than a 3-year potato history (potatoes planted three consecutive years) or whole-field plantings of golden nematode-resistant potato varieties.

Exposed fields will be surveyed at a low level of detection (50,000 or 100,000 or 200,000 cyst-per-acre level in the top 4 inches of soil). Program personnel determine the intensity of the survey and the area of implementation.

Nonexposed fields will be surveyed either mechanically or manually. Mechanical surveys on nonexposed fields will be conducted at the 500,000 cyst-per-acre level. Manual surveys on nonexposed fields will be conducted using the 8-by-8 block method, which gives a detection level of 1,000,000 cysts-per-acre.

**Do not** routinely survey formerly-infested regulated fields that are planted in accordance with New York State regulatory requirements unless the prescribed crop rotation is violated.

### Survey of Seed Potato Production Areas

All land in New York State that is used for the production of certified seed potatoes is to be surveyed annually to ensure freedom from golden nematode.

Land identified by program personnel as exposed to golden nematode infestation is to be surveyed following each crop of golden nematode susceptible potatoes provided the land has been planted a minimum of three years with susceptible potato varieties either consecutively or in rotation with other crops.

A map of the field surveyed may also be generated using Geographical Information System (GIS) software such as ArcView. With the GIS method, an outline of the field with key landmarks is created and the field crew enters information to indicate sample collection areas.

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## Soil Surveys

Soil surveys are made using manual or mechanical samplers. Prior to beginning the soil survey work, contact all growers involved in the survey to inquire and determine if the growers have encountered any trouble spots in potato or tomato fields. See [Plant Damage](#) on page 2-1-6.

Field soil sampling is used for detection and delimiting purposes. The pattern and number of samples to be taken will vary depending upon the likelihood of infestation, acreage involved, personnel available, and other factors.

Where surveying all potato or tomato fields is **not** possible, selection should be based on crop history. Fields which have been planted to potatoes and/or tomatoes consecutively for the longest period of time should be selected. There is little likelihood of finding nematode in fields planted to a host crop for less than three (3) consecutive years. If crop rotation is practiced on a field, then a longer period of time is required for the nematode to build up to a detectable level.

## Sanitation

Every reasonable precaution **must** be taken to prevent the spread of golden nematode by program personnel or equipment.

Particular care **must** be taken when working in golden nematode infested fields. Each person and piece of equipment involved in program activities of the golden nematode quarantine is a potential carrier of this pest.

This includes equipment and materials entering any golden nematode-infested field or land (such as phone company trucks and equipment, telephone company trucks and equipment, cable company, etc.). In addition, vehicles, trucks, and all other farm equipment **must** be steam cleaned before being sold or removed from any golden nematode infested farm.

### Cleaning Vehicles

Vehicles (cars, tractors, trailers, trucks, etc.) should remain on hard-surface roads. When driving vehicles on field roads is necessary, then each vehicle **must** be thoroughly sanitized as follows before leaving the survey site:

1. Remove all soil from vehicles and equipment, using power washing equipment (standard equipment for removing soil from vehicles).
2. Clean the vehicles and equipment with steam or water under pressure using a single orifice nozzle.
3. If the survey is of seed potato production areas, then additional sanitary procedures are required.
  - A. **Never** use vehicles or mechanical soil sampling equipment on seed potato production land.
  - B. **Never** use equipment used for golden nematode survey on infested or exposed land will ever be used on seed potato production land.



**No** vehicles or mechanical soil sampling equipment can be used or enter seed potato production land. **No** equipment for golden nematode survey that has been used on infested or exposed land can be used on seed potato production land.

## Cleaning and Disinfecting Survey Materials

If you survey the same field on the same side of the road (same field number), then this is considered as one field. You **do not** have to clean and bleach until leaving the field. You **must** clean after leaving the field and before starting to survey another field. If you are in a different field number, clean everything (mandatory).

### Cleaning of Clothing, Equipment, and Supplies During Survey of Commercial Potato Fields

For survey of commercial potato fields, each cleaning **must** be completed as follows before leaving one field and continuing to the next field:

1. Provide each crew member with a stiff bristle brush.
2. Each crew member will use the brush to clean all soil from trousers, shoes, and materials.
3. Thoroughly clean trowels to be used for sample collection. Trowels **must** be free of recesses or grooves where soil might become impacted.
4. Before leaving the property, clean mechanical sampling equipment with steam or water under pressure using a single orifice nozzle to remove all soil from the equipment

### Cleaning and Disinfection for Survey of Seed Potato Fields

All surveys conducted on seed potato land will include be accomplished by the following sanitation procedures:

1. Provide personnel sampling seed potato lands with lightweight neoprene boots to wear as follows:
  - A. Tuck trouser or pant legs inside these boots before entering the sampling field.
  - B. Keep pant legs tucked inside boots while samples are being collected.
2. Before entering each field and again when leaving each field, clean and disinfect all boots and equipment as follows:
  - A. Use a stiff bristle brush to remove all soil.
  - B. Scrub the boots with a solution of 5.25 percent hypochlorite in 10 parts water/

### Cleaning and Disinfection for Confirmatory and Other Surveys

Sanitary procedures outlined for seed potato fields will be followed while conducting confirmatory surveys and at any other time the inspector deems appropriate.

## Systematic Manual Soil Sampling

Upon arrival at the premises to be surveyed, the inspector should do as follows:

1. Look over the land.
2. Determine the boundaries and size and shape of the field.
3. Look at the field and determine with width and length.
4. Plan how the property should be surveyed.
5. Divide the property into a grid and record the grid on the field map.
6. Sample the field following the map grid pattern.
  - A. When completed in a uniform method, this should provide the correct soil sample size for processing at the laboratory.
  - B. If golden nematode infestation is found, then return to a specific block should be easier.



Photo by Steve Kupper

**FIGURE 2-3-1 Manual Soil Sampling**

Follow the instructions on the following pages for the specific survey type.

### 8-by-8 (8 x 8) Block Method

In the manual survey 8 x 8 Block Method, fields are divided into units of approximately 2080 square meters (half an acre), usually 32-by-112 paces (1 pace = 2-1/2 feet long).

1. Divide the field into units.
2. Determine the number of bags needed in the first tier (length of an edge of the field in a straight line), as follows
  - A. Pace the length of the field.
  - B. Divide the number of paces by 112. This gives you the number of sample bags required for each tier. If there is a remainder, then the last tier will be less than 32 paces wide.
  - C. Place the sample bags for the first tier, then take 32 paces and place the sample bags for the second tier, and continue pacing and placing bags for each consecutive tier.
  - D. If the last tier is less than 32 paces wide, then each soil sample bag for this tier will cover an area approximating the square area covered by the standard bags in the other tiers.
3. For the initial survey, sample each area in the grid pattern by collecting one scoop (or dip) of soil every eight paces and placing the soil sample in the sample bag.
4. For more intensive surveys, take one scoop or dip of soil at either 4-by-4 or 2-by-2 pace intervals.

The 8-by-8 block sampling method has 56 sample points. The soil sample bags should each contain 56 scoops of soil and weigh between 4 and 6 pounds.



Photo by Steve Kupper

**FIGURE 2-3-2 8 x 8 Sampling Method**

### Simplified 8-by-8 Block Sampling Method

The simplified 8 x 8 sampling may be employed. Rather than dividing the survey field into blocks, distribute the collection bags along the edge of the field at intervals in multiples of 8 paces, so that each will cover as close to a half-acre as possible.

**EXAMPLE** For a field approximately 450 paces long, pacing the length of the field one time (one trip) and taking a soil sample (one scoop each 8 paces) would yield one sample (8 paces per scoop = 56 scoops per sample).

The advantages of the simplified 8-by-8 Block Sampling Method are that: (1) carrying the extra weight of several soil samples while completing the last is **not** necessary; and (2) indicating the location of samples is simpler (show where the first and last samples were taken and the width between sample lines).

### Modified 8-by-8 (4-by-8) Block Sampling Method

The modified block sampling method (4 x 8) is a variation of the standard 8-by-8 grid pattern. This modified method increases the sensitivity of the survey by doubling the number of sample points, and **does not** generate any additional samples per acre.

The general procedure for the 4-by-8 modified sampling method is the same as the standard 8-by-8, except a sample is collected every 4 paces instead of every 8 paces.

1. Divide the field into units.
2. Determine the number of bags needed in the first tier (length of an edge of the field in a straight line).
  - A. Pace the length of the field.
  - B. Divide the number of paces by 112. This gives you the number of sample bags required for each tier. There may be a remainder; if so, the last tier will be less than 32 paces wide.
  - C. Place the sample bags for the first tier, then take 32 paces and place the sample bags for the second tier, and continue pacing and placing bags for each consecutive tier.
  - D. If the last tier is less than 32 paces wide, then each soil sample bag for this tier will cover an area approximating the square area covered by the standard bags in the other tiers.
3. Collect one scoop every 4 paces and place in the sample bag. Continue to maintain the 8-pace interval between sample lines.

Using the modified system, a complete sample will consist of 112 sample points.

### Mechanical (Wheel) Soil Sampling

A tractor-mounted machine is available for collecting soil samples at predetermined intervals. The tractor's operating speed is determined by field conditions for both safe tractor operation and optimum sample collection.

All mechanical equipment and probes **must** be cleaned prior to removal from the field and before entering the next field. (See [Sanitation](#) on page 2-3-4.)

Mechanical (wheel) soil sampling is taken by a tractor mounted machine that has either two or three rotating wheels with probes which penetrate the ground to a maximum depth of 4 inches. The probes are removable, and the number of probes installed determines the spacing between soil sample points. Available selection for installation is 10, 20, 40, and 80 inches between points, with 8, 4, 2, and 1 probe, respectively, per wheel. Approximately one gram of soil is collected at each point and deposited into a premarked paper bag. When possible, the swath direction will be perpendicular to normal tillage, planting, and harvest operations. See [Figure 2-3-3](#).



**FIGURE 2-3-3 Three-Wheeled Mechanical Sampler**

**Figure 2-3-4** contains a selection guide for collecting samples by machine.

Cysts per Acre <sup>1</sup>	Number of Chisels/Wheels	Swath Width (in square feet)	Area per Sample Point	Pounds of Soil Per Acre <sup>2</sup>
50,000	8	1.15	1.25	74.9
100,000	8	3	2.50	37.4
200,000	4	3	5	18.7
300,000	4	4.5	7.5	12.5
400,000	4	6	10	9.3
500,000	4	7.5	12.5	7.4
1,000,000	2	7.5	5	3.75

**FIGURE 2-3-4 Selection Guide for Collecting Soil Samples by Machine**

- 1 Detection level based on sampling from top 4 in, with vertical homogeneity of cysts assumed within the plow layer. Soil density assumed to be 86.09 lb. per square foot. Detection probability is 95 percent as determined by the Poisson approximation.
- 2 Based on one gram of soil per sample point.

---

## Selected Area Soil Sampling

For areas which are distant from known infestations and especially where large acreage is involved, the preferred method may be sampling using a selected pattern.

In the selected area soil sampling method, samples are collected from the spots most likely to be infested, as follows:

- ◆ Dumping areas for debris from graders or storage debris
- ◆ Edges where equipment is turned during cultivation and plowing
- ◆ Entrances and exits
- ◆ Low spots

Take samples in strips at intervals which are perpendicular to the direction of cultivation. The number of soil samples taken per field will depend upon the size of the field, likelihood of infestation, program objectives, and other factors.

### Peripheral Survey

Analysis of survey records from previous surveys indicates that over 90 percent of all known infestations have been recovered from a peripheral area of a field that would be encompassed within 64 paces (160 feet) from each side, and 224 paces (460 feet) from each end of the field.

In a peripheral survey sampling method, the sample collection can be either manual or mechanical. The field to be sampled **must** exceed five (5) normal 8-by-8 bags in width and six (6) normal 6-by-6 bags in width for this survey procedure to provide a significant time-saving benefit.

If the central core of the field contains a low spot that receives drainage from other parts of the field, then the central core would be tested; otherwise this core is **not** tested. \

### Grader Survey

The grader survey offers a low-cost survey method that can be used where limited resources will **not** permit a more intensive survey.



A grader survey is **not** recommended for essential early detection or delimiting purposes.

In a grader survey, standard size soil samples are collected at regular intervals during the harvest season, from soil which accumulates as potatoes are being unloaded from harvest trucks at soil facilities. Timing and frequency of soil collection depend on harvest activity, program resources, and survey objectives.

### Nursery Survey

Collecting soil samples in a nursery survey may present unique problems, especially cold frames, greenhouses, nurseries, and plant beds.

1. Divide the nursery into a grid pattern either according to type of stock grown or according to the natural boundaries (such as roads and walkways).
2. Record the grid on the map.
3. Collect soil samples and mark the sample origin on each bag
4. If collecting from piles of potting soil, take a sample from each pile and mark the location of each sample's origin on the sample bag. Survey Times and Types

### Post-crop Survey

The bulk of survey activities take place at harvest or immediately after harvest. Surveys should be carried out soon after harvest to avoid interfering with normal post-harvest farming operations.

The principal sampling procedures for post-crop survey are systematic manual soil sampling or mechanical (wheel) soil sampling.

### Rested Field Survey

If requested by the farmer(s) and if the land is to be removed from host-crop production during that year, then land normally scheduled for fall survey (post-crop) in a particular year may be sampled in the spring of the same year.

Rested field survey methods include systematic manual soil sampling and mechanical (wheel sampling).

### Confirmation Survey

PPQ's practice is to confirm initial findings of infestation by a second or confirmation survey of the property involved. This precaution is taken to preclude improper classification of properties.



The collection of additional samples (confirmation survey) is particularly important in the case of a positive find (golden nematode infestation) in a new county or State.

If multiple golden nematode cysts are recovered with a **minimum** of one viable cyst in two or more locations in areas of known infestation (such as Long Island, Livingston, Orleans, Seneca, Steuben, and Wayne counties in New York), then confirmation surveys may be omitted. However, the decision regarding the need for additional soil sampling will be left to the discretion of the appropriate Federal and State supervisory field personnel.

Preliminary identifications representing new county or State collections from Avoca, or Westhampton Beach, New York are confirmed by a nematologist in Beltsville, Maryland. Where new county or new State records are involved, a confirmation survey is **mandatory**.

### Post Resistant-Variety Treatment Survey

In a post resistant-variety treatment survey soil samples are taken from fields where two successive crop years of resistant varieties have been grown as the primary control treatment. Soil samples are collected following harvest of the second annual crop of resistant varieties.

Methods include the manual survey or the mechanical (wheel) survey. Manual survey should be done using the 4-by-4 block method. Mechanical (wheel) survey should be done at the 200,000 cyst per acre level.

All cysts **must** be nonviable. If the survey is negative and the grower has opted for the crop rotation procedure, then the field should be extensively surveyed again the first-year susceptible crop varieties are grown.

If tests confirm all cysts are nonviable after the extensive the follow-up survey, then **no** further survey needs to be conducted on this land as long as the approved rotation system is **not** violated. If there is any indication that approved cropping sequences are **not** being followed, then surveys should be resumed immediately.

## Survey to Release Land from Exposed Status



Exposed land is **not** eligible for a survey to release land from exposed status until five (5) years **after** the last viable infestation is known to have occurred on a particular grower's land and the required negative survey has been accomplished.

To establish eligibility for a survey to release land from exposed status, the exposed land **must** meet the following minimum criteria:

- ◆ Five years **must** have passed after the last viable infestation is known to have occurred on a particular grower's land; the five years are counted starting after the **required** negative survey has been accomplished
- ◆ Minimum of five host crops (potato) **must** be planted on the exposed land before the survey can be considered

A survey to release land from exposed status **must** be conducted at the 200,000 cyst per acre level or less, in the top 4 inches of soil, and **no** sooner than the fifth (5) host-crop year.

Surveys will be conducted after harvest to take advantage of the soil mixing which occurs during harvesting operations. The survey method may be manual or mechanical (wheel). If a manual survey is used, then the 4-by-4 block method or 2-by-2 block method will give the desired level of detection. All surveys **must** be negative.

Surveys for surveillance purposes may be made prior to the fifth host-crop year.

### Survey Outside Regulated Areas

Generally, conduct surveys outside of regulated areas. However, the exception is New York State where surveys will be conducted in suspect areas. A biometrics survey of major growing areas may be made periodically.

### Symptom Surveys

Symptom surveys are conducted in potato and tomato production areas throughout the United States.

Symptoms of golden nematode infestation include stunting, yellowing, and failing of crops (crop failure). Infestation symptoms occur in a spotty manner in the field. The spotty areas tend to elongate in the direction of cultivation due to the spread of golden nematode by machinery.

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## **Surveys Outside of New York State**

Conduct detection surveys on host cropland in designated suspect areas outside the State of New York. If golden nematode infestation is found, continue to follow the procedures in this manual.)

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## Sample Collection Bag Transport

The integrity of the samples **must** be maintained at all times.

### Sample Transport Vehicles

Each vehicle in which sample bags will be transported **must** be properly disinfected prior to placing the samples in the vehicle.

Program vehicles for transporting samples **must** remain on the road, highway, or thoroughfare that is near, but **not** on the infested field property.

### Seed Potato Soil Sample Transport Vehicle

Ideally, seed potato soil samples are **only** placed in a vehicle<sup>1</sup> that has **never** been in infested areas or carried samples from infested areas.

If a program does not yet have a vehicle dedicated to seed potato soil sampling transport only, then another vehicle may be used provided the interior and exterior has been completely disinfected with steam heat treatment. Loading Sample Collection Bags

The survey crew will place the soil samples into the program vehicles. Transport vehicles **must** remain off the survey property, and on the highway, road, or thoroughfare.

The survey crew will load the sample bags into the transport vehicle (van or truck) as follows:

1. Keep sample collections separate. Use cardboard or empty space between the collections to prevent commingling.
2. Place each sample bag in the vehicle in consecutive order, by the bag number, either starting with the first bag (1) and ending with the last bag, or starting with the last bag and ending with the first bag (1).
3. If there are bags from more than one collection, then keep each collection separate, and individually place the bags in consecutive order as stated above, on one side of the vehicle.

<b>EXAMPLE</b>	All sample bags in the collection labeled with red marker are stacked on the left. All sample bags in the second collection labeled with blue marker are stacked on the right. A piece of cardboard is between the collection.
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4. To prevent commingling of the collections, either place a piece of cardboard between each collection or keep an empty space between each collection.
5. The soil samples are then transported to the Golden Nematode Laboratory in Avoca, New York for examination.

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1 Currently, one truck in Avoca is dedicated for seed sample collection transportation.



Photo by Steve Kupper

**FIGURE 2-3-5 Single Soil Samples and Supplies Loaded in Van for Transport to the Work Unit Laboratory**



# 2

Golden Nematode  
Program Manual

## Procedures

### *Laboratory, Rack Room, and Wash Room*

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#### Contents

Introduction	page 2-4-1
Work Unit	page 2-4-2
Rack Room	page 2-4-2
Step 1: Unload the Vehicles	page 2-4-3
Step 2: Disinfect and Restock Transport Vehicles	page 2-4-4
Step 3: Allow the Samples to Dry	page 2-4-5
Step 4: Gather a Sample Collection from the Rack Room	page 2-4-6
Sample Wash Room	page 2-4-9
Step 1: Gather Materials for Sample Washing	page 2-4-10
Step 2: Prepare Cleaning Solution	page 2-4-10
Step 3: Wash Hands and Wear Proper Attire	page 2-4-11
Step 4: Place Sample Bags and Beakers for Sample Washing	page 2-4-12
Step 5: Wash Mineral Soil Samples	page 2-4-14
Step 6: Process Muck Soil Samples	page 2-4-16
Step 7: Place Beakers on Laboratory Counter	page 2-4-17
Step 8: Clean and Disinfect the Sample Washroom	page 2-4-18
Laboratory Sample Reading	page 2-4-19
Step 1: Set Up the Sample Reading Station	page 2-4-20
Step 2: Set Up the Golden Nematode Cyst Station	page 2-4-21
Step 3: Prepare for Sample Reading	page 2-4-21
Step 4: Read the Samples	page 2-4-24
Step 5: Remove Cysts	page 2-4-26
Golden Nematode Cyst Identification	page 2-4-28
Record of Infestation Folder	page 2-4-28

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#### Introduction

The *Laboratory, Rack Room, Wash Room* section provides the work unit procedures for unloading and stacking soil sample bags, washing the soil samples, examining the samples, and preparing suspect cysts for positive identification.

## Work Unit

Each sample passes through the following three areas within the Work Unit:

- ◆ Rack room
- ◆ Wash room
- ◆ Laboratory

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## Rack Room

The rack room is climate-controlled with a dehumidifier and heater to assist with drying soil sample bags. Up to 10,000 sample bags can be dried and stored in this room until moved to the wash room for sample washing. Racks and shelves are each labeled with a rack letter and sample bag number. See [Figure 2-4-1](#) below.



**FIGURE 2-4-1** Rack Number and Shelf Number Label

Continue to [Step 1: Unload the Vehicles](#) on page 2-4-3.

## Step 1: Unload the Vehicles



Maintain the integrity of each soil sample by keeping the bags in consecutive order by sample bag number.

Upon return to the USDA-APHIS-PPQ Work Unit facility from the sampling field, the crew will park the transport vehicle(s) (vans/trucks) in a row, and unload the sample bags as follows:

1. Place the safety ladder at the racks where the sample bags will be placed.
2. Crew members will line-up single file from the vehicle containing the sample bags to the laboratory rack room.
3. Start unloading the highest-numbered bag (collection end bag), and pass each bag down the line. Keep the collection together and in sequential order as the bags are being unloaded.
4. A crew member places the collection end bag at the front of the rack shelf, and stacks each bag in consecutive order behind the collection end bag.
5. The Crew Leader records on the *Sample Storage* sheet, the date the collection is placed in the rack room, the collection bag number, the total number of samples in the collection, the sample bag placement rack number, and the shelf number(s). (See [Sample Storage Sheet](#) on page A-1-13.) The information on this sheet is useful to identify the location of each sample bag while in the rack room.

## **Step 2: Disinfect and Restock Transport Vehicles**

After all bags have been unloaded and placed in the rack room, then disinfect all vehicles used to transport samples from the field to the laboratory facility, as follows:

- 1.** Move every vehicle used to transport samples from the field, away from the building.
- 2.** Remove all supplies from the vehicle.
- 3.** Vacuum vehicle interior (carpet, seats, sides, ceiling, floor, floor mats, and rear area).
- 4.** Prepare a bucket of bleach water solution one part 5.25 percent hypochlorite to 10 parts water.
- 5.** Wash entire interior with bleach water solution and sponge to disinfect the interior.
- 6.** The vehicle must be cleaned, disinfected, and spotless at the end of each trip from the field to avoid contamination, even when the vehicle will be driven back to the same field for sampling the next day.
- 7.** Wash and disinfect the supplies and equipment.
- 8.** Reload the sampling supplies and equipment into the transport vehicle.
- 9.** Clean and disinfect the exterior of the water jugs and fill with water for the next day.
- 10.** If a van is used to transport samples, then stop at a car wash on the return trip back to the Work Unit facility.

### Step 3: Allow the Samples to Dry

Sample bags remain on the racks in the drying rack room for approximately two months to dry depending on the moisture content of the soil at the time of collection. Adjust the dehumidifier and thermostat as rack room humidity and temperature levels dictate. When heat is used, the temperature should usually be 60°-65°F.



**FIGURE 2-4-2 Soil Samples Drying in the Rack Room**

Muck soil is generally friable. Mineral soil dries much more slowly and is more susceptible to hardening. The soil and potential cysts must be dry to process.

### Sample Moisture Content

Determine if the soil is dry enough for sample washing, as follows:

1. If the bag still feels heavy, this could indicate the soil is still too moist.
2. Check the outside of the bag for wrinkled or damp or wet spots. If found, this indicates the soil is too moist; return the bag to shelf (exact spot where taken from).
3. If the bag is completely dry, then remove the staples from a bag and unfold the top,
4. Look inside the bag for any moist soil sticking together or any dark spots in the soil due to moisture content. If found, staple the bag shut and return to the shelf.

## Sample Clumps or Hardening

After you confirm the sample bag contents are dry, feel the bag to determine if the soil has hardened or has clumps. The soil **must** be loose to process in the sample washroom. To break up the samples, do as follows:

1. Place the sample bag inside a 4-mil plastic bag, and secure closed to avoid contamination.
2. Take the sample and place on the concrete floor.
3. Take a rubber mallet and pound the bag to break up all the clumps and chunks of soil.

### Step 4: Gather a Sample Collection from the Rack Room

After sample bags in the collection have cured (dried) on the racks, they are ready for processing. Two Biological Aides or Biological Laboratory Technicians are needed to gather the sample bags and record numbers; one to remove the bags from the shelves, and another to assign and record beaker and sample numbers, and stack the bags onto the appropriate cart.

1. Gather the following materials:
  - ❖ Carts, flat-surface, each a different color (one red and one blue or one red and one purple), **no** hooks (2)
  - ❖ Clipboards (2)
  - ❖ Markers, permanent, two colors (one red and one blue or one red and one purple) (2)
  - ❖ Safety ladder
  - ❖ *Sample Storage* sheets (from rack)
  - ❖ *Golden Nematode Laboratory Sample Processing Daily* sheets



You **must** use a one color permanent marker for the first collection and a different for the second collection to help keep the collections separate during processing.

2. If you need to remove sample bags from a high rack, then place the safety ladder at the rack and climb the ladder to remove the bags.
3. Place a cart (red), a marker (red) and a clipboard with the *Golden Nematode Laboratory Sample Processing Daily* sheet (already completed by the Laboratory Leader for the collection to be removed) near the shelves.
4. Go to the shelf, look under the BEAKER NO. column on the *Golden Nematode Laboratory Sample Processing Daily* sheet and verify the collection number listed on the form is the sample as the collection number on the bag.

5. If the collection "END" bag (just above the bag number in the lower left-hand corner) is at the front of the rack shelf, then remove this bag first and hand to a second person.
6. The second person will locate the following information on the Golden Nematode Laboratory Daily sheet and verify each number matches the sample bag label:
  - A. Collection number (under BEAKER NO.)
  - B. Beaker number (under BEAKER NO.)
  - C. Bag number (under BAG NO.)
7. Write the pre-assigned beaker number in the center of the bag. See [Figure 2-4-3](#) below.



**FIGURE 2-4-3** Sample Bag Beaker Number (50), Collection Number (DEM-03), Field Name (38-B-40), Bag Number (24), and Collection Date (1/11)

8. Place the bag in consecutive order by Beaker No. on the red cart, and repeat.
9. Stack the bags on the cart in consecutive order or reverse consecutive order.



Photos by Dan Kepich

**FIGURE 2-4-4 Beaker Numbers to Sample Bags**

- 10.** If time allows after you gather the first collection, then get a second cart (blue or purple) and repeat 1., through 8., above, **except** use the blue or purple marker and cart. **Do not** use red for the second collection.
  - A.** Use the blue or purple marker to number the second batch of sample bags.
  - B.** After the beaker number is written on the bag, then place the second collection on the blue cart.

## Sample Wash Room

Sample processing is conducted daily. **Do not** wash more samples than can be read within three hours of washing on the same day. Soil samples are washed using Fenwick Can Washers (soil sample washing machine). See [Figure 2-4-5](#) below.



Photo by Dan Kepich

**FIGURE 2-4-5 Fenwick Can Washers (Soil Sample Washing Machine) in Sample Wash Room**

### Step 1: Gather Materials for Sample Washing

Gather the following materials for sample washing:

- ◆ Beakers, numbered 1-83, empty
- ◆ Bleach (hypochlorite)
- ◆ Bucket and mop
- ◆ Carts (2 flat surface, no hooks; each cart **must** be a different color)
- ◆ Lab sheet
- ◆ Pollen mask
- ◆ Respirator
- ◆ Rubber apron
- ◆ Rubber gloves
- ◆ Safety glasses
- ◆ Sieve with 20 mesh screen (top sieve)
- ◆ Sieve with 60 mesh screen (bottom sieve)
- ◆ Tank

There are two tanks for sample washing, so two people can process samples at a time. Be careful to keep the collections separate.



Be careful **not** to cross-contaminate the samples.

### Step 2: Prepare Cleaning Solution

First thing each morning, prepare a disinfectant solution by mixing 1 cup bleach to 3 gallon of hot water in the bucket. You will need this to mop the floor during the day.

### Step 3: Wash Hands and Wear Proper Attire

Wash your hands or wash off your gloves as follows:

- ◆ Before you begin sample washing
- ◆ Before putting on rubber gloves
- ◆ After opening each sample bag
- ◆ After pushing sample bags down into trash bag
- ◆ After pushing the trash bag down into the trash can
- ◆ After closing and removing the trash bag from the trash can and wash room

Put the pollen mask, rubber apron, rubber gloves, safety glasses on before entering the laboratory wash room.



Some people choose **not** to wear rubber gloves during processing, due to sensitivity. If you **do not** wear gloves, then be sure to wash your hands between sample washing.

### Step 4: Place Sample Bags and Beakers for Sample Washing

Remove sample bags from the cart in groups of 25, in consecutive order and place in consecutive order on the wash room counter.

<b>EXAMPLE</b>	Remove sample bags numbered 1-25 from the cart, and place in consecutive order on the counter.
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1. Carefully slice the top of each bag open with a knife, but **do not** dig down into the sample soil. Be especially careful when opening muck soil sample bags as the soil is very dry and can be dusty.



**FIGURE 2-4-6** Opening Sample Bags

2. Wash your hands, but **do not** hose them clean, between opening the sample bags and before picking up the beakers.

3. Select the beaker number which corresponds with the sample bag number and confirm that both the sample bag number and the beaker number are identical. See [Figure 2-4-7](#) below.



**FIGURE 2-4-7 Beaker Number Confirmation**

4. Take both the sample bag and the beaker to the can washer. To wash mineral soil samples, continue to [Step 5: Wash Mineral Soil Samples](#) on page 2-4-14. To wash muck soil samples, continue to [Step 6: Process Muck Soil Samples](#) on page 2-4-16.

### Step 5: Wash Mineral Soil Samples

To wash mineral soil samples, do as follows:

1. Turn the low water on and place the hose in the tank with the spout, so there will be a little water in the tank. See the large tank on the right in [Figure 2-4-8](#) below.
2. Open soil sample bag, and dump slowly into tank and turn high water on while dumping sample.



**FIGURE 2-4-8 Tank and Dumping Sample**

3. Allow high water to flow just up to the lip of the tank, then turn the high water off; this brings the soil up and gets everything to the top of the tank.
4. Keep the low water flowing all through the washing process.
5. Discard empty sample bag into trash can.

6. As the debris comes up to the top, carefully hose and skim the larger debris from the tank into the top sieve (first sieve; 60 mesh screen). See [Figure 2-4-9](#) below.



**FIGURE 2-4-9 Sieves and Skimming Debris**

7. As the larger debris is skimmed off with the running water, the golden nematode cysts will float and come right up to the top (you **cannot** see the cysts with the naked eye). Gently skim off the nematode cysts with smaller debris into the first (top) sieve (60 mesh screen)
8. Thoroughly hose down the top screen and sieve sides to force everything remaining in the top sieve down into the lower sieve (bottom screen). This forces the flotsam down into the bottom screen (100 mesh screen).
9. Thoroughly hose bottom screen and force all material (flotsam and cysts) from the bottom sieve into a beaker keep adding water to the screen until level is about 2" from the beaker's top.
10. Take the bottom sieve, and carefully hose the flotsam from the bottom screen into the same-numbered beaker.
11. Take the hose and add clear water into the beaker until about 2" from top.
12. Take the top sieve into the sink and thoroughly clean before placing back into tank.
13. Wash your hands; **do not** hose them clean.

When processing mineral soil samples **only**, you can wash one sample and then start another in the second tank. While the first tank is filling with water, get a second sample bag and begin processing in the second tank. **Do not** mix the contents of sample bags together.

### Step 6: Process Muck Soil Samples

Washing muck soil samples is much more time-consuming due to the extremely dry nature of the soil,

1. Wear a pollen mask, respirator, and rubber apron, when processing muck soil samples.
2. Process **only** one sample at a time.
3. Keep water on low **only** during washing; **do not** turn water on high water.
4. Turn the low water on and place the hose in the tank with the spout, so there will be a little water in the tank. See the large tank on the right in [Figure 2-4-8 on page 2-14](#).
5. Open the soil sample bag carefully, and slowly dump the sample into tank; keep water on low while dumping sample.
6. Be extremely careful. Muck soil can resist wetting.
7. Hold the high-pressure hose and slowly break up the soil.
8. Allow low water level to flow just up to the lip of the tank; this brings the soil up and gets everything to the top of the tank.
9. Keep the low water flowing all through the washing process.
10. As the debris comes up to the top, carefully hose and skim the larger debris from the tank into the top sieve (first sieve; 60 mesh screen). See [Figure 2-4-9 on page 2-15](#).
11. As the larger debris is skimmed off with the running water, the golden nematode cysts will float and come right up to the top (you **cannot** see the cysts with the naked eye). Gently skim off the nematode cysts with smaller debris into the first (top) sieve (60 mesh screen)
12. Thoroughly hose down the top screen and sieve sides to force everything remaining in the top sieve down into the lower sieve (bottom screen). This forces the flotsam down into the bottom screen (100 mesh screen).
13. Thoroughly hose bottom screen and force all material (flotsam and cysts) from the bottom sieve into a beaker keep adding water to the screen until level is about 2" from the beaker's top.
14. Take the bottom sieve, and carefully hose the flotsam from the bottom screen into the same-numbered beaker.
15. Take the hose and add clear water into the beaker until about 2" from top.
16. Take top screen into the sink and thoroughly clean before placing back into tank.
17. Discard the empty sample bag into a wash room trash can.

### Step 7: Place Beakers on Laboratory Counter

Beakers numbers 1-83, are assigned to help prevent the commingling of samples and to keep track of the sample processing in the lab.

1. As each sample is washed and poured into the correct beaker, take beaker and place on cart.
2. Line up the beakers by number in consecutive order on the laboratory counter.



**FIGURE 2-4-10** Sample Beakers on Laboratory Counter

After all sample washing is completed, continue to **Step 8: Clean and Disinfect the Sample Washroom** on page 2-4-18.

### **Step 8: Clean and Disinfect the Sample Washroom**

Thoroughly clean and disinfect the washroom and carts (after each collection is processed), as follows:

- 1.** Remove the rubber apron and hose off in the sink, then wash the apron.
- 2.** Hose off the entire wash area. Use a clean sponge to wipe down the hood and screens, and the inside and outside of sinks.
- 3.** Make sure there is **no** dirt left on the sides and in the bottom of the sinks. Hose off the sinks again to be sure.
- 4.** Remove all dirt from the counters.
- 5.** Wash the countertops and backsplash with bleach solution.
- 6.** Sweep the washroom floor.
- 7.** Wash the washroom floor with mop and the bleach solution.
- 8.** Wash the carts with a sponge.
- 9.** Wash your hands thoroughly.
- 10.** If there is another batch of samples to process, then start at the beginning and process the next batch of samples.
- 11.** If sample washing is finished, then begin sample reading.

Continue to ***Laboratory Sample Reading*** on page 2-4-19.

## Laboratory Sample Reading

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Everything used in the laboratory **must** be clean to avoid contaminating the samples. Keep the work space clean and thoroughly clean the laboratory when finished.

### Materials Needed

Gather the following materials:

- ◆ Ball point pen or thin permanent marker
- ◆ Empty beakers, 600 mil
- ◆ Daily Sample Processing Sheet
- ◆ Dissecting microscope
- ◆ Glass vials (10 dram) with lids
- ◆ Muffin tin sieve
- ◆ Plastic muffin tin sieve holder
- ◆ Probe
- ◆ Sample beakers, 600 mil
- ◆ Individual microscope light
- ◆ Spatula
- ◆ Stainless steel click counter
- ◆ 3-section plastic tray
- ◆ Squirt water bottle with spout
- ◆ Vial labels
- ◆ Water
- ◆ White lab coats

### **Step 1: Set Up the Sample Reading Station**

Gather the following materials for each golden nematode sample reading station:

- ◆ Beaker, 600 mil, empty
- ◆ Dissecting microscope
- ◆ Loop
- ◆ Probe
- ◆ Scope light, individual
- ◆ Squirt water bottle



Photo by Dan Kepich

**FIGURE 2-4-11 Sample Reading Station**

## Step 2: Set Up the Golden Nematode Cyst Station

Gather the following materials and for the golden nematode station:

- ◆ 10 dram glass vials with lids
- ◆ Specialty microscope slide labels
- ◆ Dental loop
- ◆ Pen, ball point or thin tip permanent marker
- ◆ Scotch tape



FIGURE 2-4-12 Golden Nematode Station

## Step 3: Prepare for Sample Reading

Prepare for sample for reading as follows:

1. Put on a clean, white laboratory coat.
2. Place a 100-mesh muffin tin sieve at the sink.
3. Take the first sequentially-numbered beaker sample from the counter.



Important

Samples **must** be read within three hours of washing. After about 3 hours, cysts and floatsam **no** longer float, and sink to the beaker bottom.

4. Go to the *Golden Nematode Laboratory Sample Processing Daily* sheet, and locate (under BEAKER NO.), the collection number and the pre-assigned beaker number you will be reading. Place your initials on the sheet beside the beaker number. See [Figure 2-4-13](#).

5. Keep the sheet on the clipboard, but place underneath the blank sheets. (The Laboratory Leader collects the completed sheets weekly.)



**FIGURE 2-4-13 Golden Nematode Laboratory Sample Processing Daily Sheet**

6. Take the beaker sample to the sink, slowly swirl the beaker sample, and pour the sample into muffin tin sieve; **do not** the empty sieve yet.

7. Add water from sink spigot into beaker, swirl the beaker water, and pour into same muffin tin again to make sure all flotsam and possible golden nematode cysts go into the sieve.



**FIGURE 2-4-14** Pouring Sampling Material from Beaker into Muffin Tin Sieve

8. Use the spatula to scrape the sample from the muffin tin sieve into the 3-section plastic tray.
9. Take the same sample and muffin tin sieve by using the squirt bottle to squirt water into the sieve to move any remaining sample material from the sieve and into the 3-section plastic tray.
10. Wash the laboratory counter and the sink with a clean sponge and water.

### Step 4: Read the Samples

Read the samples as follows:

1. Take the 3-section tray with sampling material over to the GN reading station.
2. Place the tray under the microscope.
3. Use the squirt bottle and slowly add water to the sample material in each section of the tray so that the golden nematode cysts will float to the top, but **not** flow over. All cysts will float, whether viable (live) or non-viable (old, flattened, or dead).
4. Look into the microscope, and move the plastic sample tray around until just a little bit of white plastic is showing at the upper left top.
5. Start in the upper left corner, and use the probe to move the sample material around to examine the sample.



Photos by Dan Kepich

**FIGURE 2-4-15 Flotsam Examination for Cysts**

6. Move the tray in small increments so that you see the top white middle portion of the tray. Use probe to move material around and examine the sample. Continue moving the tray and probing the material until you have examined the entire sample in each section of the tray.
7. Look for round, spherical object that appear to have a small spout or protrusion. Viable (live) cysts are usually golden; however, cysts that have survived treatment could be

dark-colored, but still be viable. Non-viable (dead) cysts are generally black. See the microscopic view of golden nematode cysts in [Figure 2-4-16](#) below.



Photo by Zaphar Handoo

**FIGURE 2-4-16 Golden Nematode Cysts**

8. If you locate a golden nematode cyst, then continue to the instructions for removing cysts [Step 5: Remove Cysts](#).
9. After you are finished examining the sample and have removed the suspect golden nematode cysts for identification, then dump the flotsam from the tray into the empty beaker.
10. Take the muffin tin sieve, plastic tray, and probe to the laboratory sink. Run water and scrub muffin tin sieve with toothbrush and soap. Check and make sure all material is removed. Rinse probe and beaker.
11. Repeat each step above until all beaker samples have been examined.

### Step 5: Remove Cysts

All golden cysts **must** be identified, whether considered to be viable or non-viable. If you locate one or more golden nematode cysts when reading a sample, then do as follows:

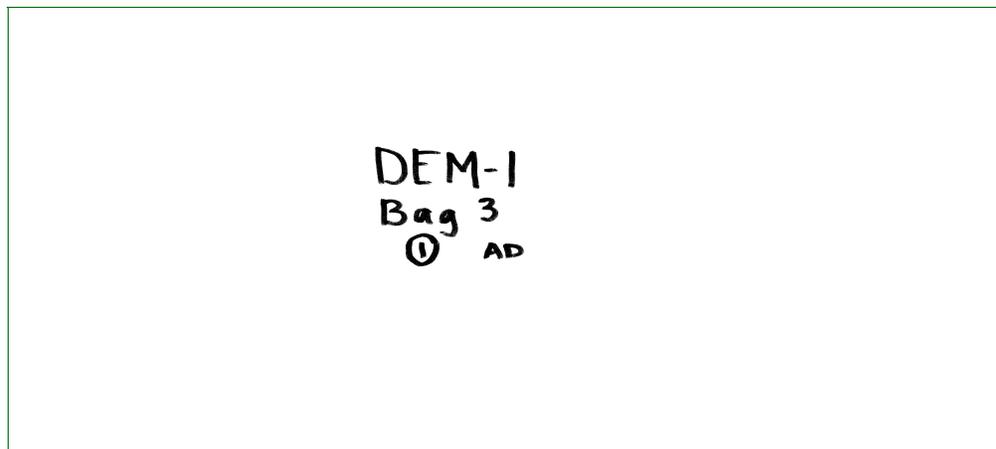
1. Get one vial and a lid from the golden nematode station and take to the sampling station.
2. Use **only** one vial per sample, even if there are multiple cysts from the same sample.
3. **Do not** mix cysts from multiple samples into one vial.
4. Use the loop, and look under the microscope to locate and isolate each golden nematode cyst.
5. Use the loop to remove the cyst(s) from the flotsam in the tray, place each cyst from the sample into the vial, and screw on the lid. See [Figure 2-4-17](#) below.



Photo by Dan Kepich

**FIGURE 2-4-17 Golden Nematode Cyst Placement in Vial**

5. Prepare a label and use a ballpoint pen or thin permanent marker to write the collection number, sample bag number, number of cysts found in the individual sample, and put your initials on the label.



**FIGURE 2-4-18** Example of Vial Label

6. Attach the label to the sample vial and cover the label with scotch tape.
7. Keep all sample vials from the collection together (**do not** mix collections).
8. Go to the *Golden Nematode Laboratory Sample Processing Daily* sheet, locate the BEAKER NO. (cysts were in) and record under COLL. NO. the number of cysts found.
9. Give the vial collection to the Laboratory Leader.

## Golden Nematode Cyst Identification

If a golden nematode identifier is available on site, then give the collection vials to the identifier.

If an on-site identifier is **not** available, then the Director or designee will do as follows

1. Complete *PPQ Form 391, Specimens for Determination*.
2. Pack the vials in packing material and place in a sturdy parcel to prevent breakage in transit and danger of pest dissemination. and enclose the completed Form 391.
3. Attach a label identifying the contents to the outside of the parcel, and ship using overnight delivery to the following address:

Dr. Zaphar Handoo  
USDA-ARS Nematology Laboratory  
Bldg. 011A, Room 159 BARC-West  
10300 Baltimore Avenue  
Beltsville, MD 20705-2350  
Phone: 301/504-6666

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## Record of Infestation Folder

When a property is found to be infested with golden nematode, the Laboratory Leader will prepare a Record of Infestation Folder original and maintain the file at the Work Unit Office. The Laboratory Leader will prepare a duplicate Record of Infestation Folder and give the duplicate folder to the Plant Health Safeguarding Specialist in charge of the regulated area. Maintain the duplicate folder in the field office.

Maintain both the **original** folder and the duplicate field office folder as the historic record of the property and safeguard as such.

Fasten the following information inside **both** the original Work Unit folder and the field office folder:

- ◆ Maps
- ◆ PPQ Form 391, Specimen for Determination
- ◆ PPQ Form 312, Golden Nematode Survey
- ◆ Records of finds
- ◆ Records of **no** finds
- ◆ Records from previous surveys on the property
- ◆ Other important information

# 3

Golden Nematode  
Program Manual

## Procedures

### *Regulatory Treatment and Certification*

#### Contents

Introduction	<b>page 3-1-2</b>
Regulated Articles	<b>page 3-1-2</b>
Special Procedures	<b>page 3-1-4</b>
Movement Under Limited Permit to Approved Destinations	<b>page 3-1-4</b>
Shipment to Mexico or Canada	<b>page 3-1-4</b>
Shipment to Foreign Countries	<b>page 3-1-4</b>
Certificates and Permits	<b>page 3-1-5</b>
PPQ Form 519, Compliance Agreement	<b>page 3-1-5</b>
<b>page 3-1-7</b>	
PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantines	<b>page 3-1-6</b>
Approved Containers and Equipment	<b>page 3-1-16</b>
Used Containers and Other Used Equipment	<b>page 3-1-16</b>
Sanitation and Treatment	<b>page 3-1-18</b>
Sanitation	<b>page 3-1-19</b>
Authorized Pesticides	<b>page 3-1-19</b>
Certification Period	<b>page 3-1-19</b>
Approved Treatments	<b>page 3-1-20</b>
Irish Potatoes, Grade A or Grade B (Except Seed Potatoes)	<b>page 3-1-20</b>
Movement of Potatoes Under Limited Permit (Except Seed Potatoes)	<b>page 3-1-20</b>
Ear Corn With Shucks Attached	<b>page 3-1-21</b>
Hay, Straw, and Plant Litter	<b>page 3-1-21</b>
Plants With Roots, Bulbs, Corms, Rhizomes, Ornamental Plant Tubers, and Sod (Other than Tomato and Eggplant Transplants)	<b>page 3-1-21</b>
Root Crops (Other than Irish Potatoes and Sugar Beets)	<b>page 3-1-21</b>
Sugar Beets	<b>page 3-1-22</b>
Small Grains	<b>page 3-1-22</b>
Soybeans (Other than for Seed)	<b>page 3-1-22</b>
Soil	<b>page 3-1-22</b>
Dry Heat Fumigation of Soil Samples	<b>page 3-1-23</b>
Steam Fumigation for Bench and Potting Soil	<b>page 3-1-24</b>
Tomato and Eggplant Transplants	<b>page 3-1-24</b>
Used Farm Equipment, Mechanized Soil Moving Equipment, Used Containers, Used Farm Tools, and Other Similar Articles	<b>page 3-1-25</b>
Inspection of Grader Stations	<b>page 3-1-26</b>
Guidelines for Releasing Land from Regulation	<b>page 3-1-27</b>
Category 1 Non-agricultural Land (20-year Status)	<b>page 3-1-27</b>
Category 2 Non-agricultural Land (Less than 20-year Status)	<b>page 3-1-27</b>
Category 3 Agricultural Land	<b>page 3-1-27</b>

## Introduction

The *Regulatory Treatment and Certification* section provides a list of the regulated articles in golden nematode quarantined areas, the conditions under which these regulated articles can be moved from the site of origin, the required certificate or permit that **must** accompany the regulated item for movement, and the procedures that the PPQ Officers **must** follow to authorize movement of the regulated articles, and guidelines for releasing land from regulation. All regulatory procedures apply to both GN Ro1 and GN Ro2.

## Regulated Articles

The following regulated articles require a certificate or permit year round:

- ◆ Compost, decomposed manure, humus, muck, peat, and soil separately or with other things



Important

**Exemption** to regulation: Soil samples shipped to approved laboratories **do not** require attachment of a certificate or permit. See the inspector for a list of approved laboratories.

- ◆ Ear corn, unshucked (**does not include** shucked ear corn)



Important

**Exemption** to regulation: If the unshucked ear corn has been harvested in bulk or directly into approved containers, and **not** exposed to infestation after cleaning or other prescribed handling, and if the corn and containers thereof have **not** come into contact with the soil, then unshucked ear corn is exempt from regulation unless notified otherwise by the inspector.

- ◆ Grass sod
- ◆ Hay, straw, fodder, and plant litter of any kind



Important

**Exception** to regulation: If hay, straw, fodder, and plant litter of any kind is moved in approved containers and **not** exposed to infestation after cleaning or other prescribed handling, then hay, straw, fodder, and plant litter are exempt from regulation unless notified otherwise by the inspector.

- ◆ Irish potatoes and other root crops

- ❖ Irish potatoes



**Exception** to regulation: If Irish potatoes are graded at an approved grader or washed free of soil, packaged in approved containers, and **not** exposed to infestation after cleaning or other prescribed handling, then Irish potatoes **other than** for seed are exempt unless otherwise notified by an inspector.

- ❖ Root crops **other than** Irish potatoes and sugar beets



**Exception** to regulation: If root crops other than Irish potatoes and sugar beets are moved in approved containers and **not** exposed to infestation after cleaning or other prescribed handling, then root crops **other than** Irish potatoes and sugar beets are exempt unless otherwise notified by an inspector.

- ◆ Plant crowns and roots for propagation

- ◆ Plants with roots (**does not** include soil-free aquatic plants)

- ◆ Small grains and soybeans

- ❖ Small grains



**Exemption** to regulation: If small grains have been harvested in bulk or directly into approved containers, if cleaned to meet State seed sales requirements, and **not** exposed to infestation after cleaning or other prescribed handling and the containers have **not** come into contact with the soil, then small grains are exempt unless otherwise notified by an inspector.

- ❖ Soybeans **other than** for seed



**Exemption** to regulation: If soybeans **other than** for seed have been harvested in bulk or directly into approved containers and the soybeans **not** exposed to infestation after cleaning or other prescribed handling and containers have **not** come into contact with the soil, then soybeans **other than** for seed are exempt, unless otherwise notified by an inspector.

- ◆ True bulbs, corms, rhizomes, and tubers of ornamental plants

- ◆ Used crates, boxes, burlap bags, and other used farm product containers

- ◆ Used farm tools

- ◆ Used mechanized cultivating and used harvesting equipment

- ◆ Used mechanized soil-moving equipment

- ◆ Any other products, articles, or means of conveyance, of any character whatsoever, **not** covered by the above, and determined by an inspector that they present a hazard for spread of golden nematode, and the person in possession thereof has been so notified

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## Special Procedures

### Movement Under Limited Permit to Approved Destinations

#### Movement Within Quarantined States

Movement under limited permit (PPQ Form 530, Limited Permit) by PPQ to destinations within quarantined States **must** be approved by the appropriate State regulatory official or his designee.

#### Movement Outside Quarantined States

Movement under limited permit (PPQ Form 530, Limited Permit) to destinations in States **other than** the quarantined States **must** be approved by the PPQ Deputy Administrator or his designee in concurrence with the receiving State regulatory officials. Shipment to Offshore States, Territories, or Possessions of the United States

Shipments of regulated articles to offshore States, territories, or possessions of the United States shall be in accordance with the procedures outlined in this manual.

### Shipment to Mexico or Canada

Shipment of regulated articles to Mexico or Canada shall be in accordance with the procedures outlined in this manual and any other requirement which may be specified by officials of Mexico or Canada.

### Shipment to Foreign Countries

Shipment of regulated articles destined to other countries shall be made in accordance with import requirements of the destination country.

## Certificates and Permits

### PPQ Form 519, Compliance Agreement

Compliance agreements can be entered with persons who grow, handle, move or sell regulated land or articles on or from regulated land.

To enter a compliance agreement, the person **must** do the following:

- ◆ Review with an inspector each stipulation of the compliance agreement
- ◆ Agree to the terms of the agreement
- ◆ Follow the terms of the agreement
- ◆ Sign the agreement

Examples of persons who would enter a compliance agreement with to have their equipment that enters golden nematode infested land or any land that is regulated for golden nematode cleaned. These would include the following:

- ◆ Auction houses and auctioneers
  - ❖ Compliance agreement to contact USDA of sale, especially of equipment used on GN land
  - ❖ PPQ Offices/PHSS will go over the consignor's list before the auction, and treat the items to be sold (equipment cannot be move before treated)
- ◆ Cable company
- ◆ Companies that apply fertilizer or lime on a regulated field
- ◆ Electric company
- ◆ Gas company
- ◆ Phone companies
- ◆ Towns and municipalities
- ◆ Miscellaneous (such as a company that is installing wind towers on GN infested land)

An inspector may cancel a compliance agreement orally or in writing whenever the inspector determines that the person who has entered into the agreement has **not** complied with the conditions of the agreement.

The inspector should review each compliance agreement on a yearly basis and update, modify, or remove as needed. Compliance agreements are valid for one year from the date of signing; a new compliance agreement **must** be signed every year.

See **PPQ Form 519, Compliance Agreement** on page A-1-29.

### **PPQ Form 530, Limited Permit**

PPQ Form 530, Limited Permit, is used to authorize movement of noncertified, regulated material to a specific approved destination for processing or treatment. See **PPQ Form 530, Limited Permit** on page A-1-35 more information.



Movement of noncertified regulated articles **must** be approved by the appropriate State Regulatory Official in the applicable State (if movement is within the quarantined State) and/or PPQ Deputy Administrator or designee and the receiving State regulatory official (if movement is outside the quarantined State).

Continue to **Table 3-1-1**.

### **PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantines**

PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantines, is used to certify specific regulated articles that are certified free from Golden Nematode. See **PPQ Form 540, Certificate of Federal/State Domestic Quarantines** on page A-1-37 for more information.

**TABLE 3-1-1 Determine if Certificate or Permit Is Required for Regulated Articles**

<b>If the article is:</b>	<b>And:</b>	<b>Then:</b>
<ul style="list-style-type: none"> <li>◆ Bulbs (true)</li> <li>◆ Corms</li> <li>◆ Rhizomes</li> <li>◆ Tubers of ornamental plants</li> </ul>	Certified free from golden nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.J.kepich@aphis.usda.gov
Compost, separate or mixed with other things	Certified free from golden nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.J.kepich@aphis.usda.gov

**TABLE 3-1-1 Determine if Certificate or Permit Is Required for Regulated Articles (continued)**

<b>If the article is:</b>	<b>And:</b>	<b>Then:</b>
Corn, ear shucked	Certified free from golden nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Corn, ear <b>not</b> shucked (unshucked)	<ul style="list-style-type: none"> <li>◆ Harvested in bulk or directly into approved containers (see <b>Authorized Pesticides on page 3-1-19</b>)</li> <li>◆ Corn and containers have <b>not</b> come into contact with the soil</li> <li>◆ <b>Not</b> exposed to infestation after cleaning or other prescribed treatment and other prescribed handling</li> </ul>	EXEMPT from regulation
	<b>Not</b> as described in the cell immediately above	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Fodder	<ul style="list-style-type: none"> <li>◆ Moved in approved containers</li> <li>◆ <b>Not</b> exposed to infestation after cleaning or other prescribed handling</li> </ul>	EXEMPT from regulation unless notified otherwise by inspector
	<b>Not</b> in approved containers or has been exposed to infestation after cleaning or prescribed handling	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov

**TABLE 3-1-1 Determine if Certificate or Permit Is Required for Regulated Articles (continued)**

If the article is:	And:	Then:
Grass sod	Certified free from Golden Nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Grains, small	<ul style="list-style-type: none"> <li>◆ Harvested in bulk or directly into approved containers and have <b>not</b> come into contact with the soil after harvesting</li> <li>◆ <b>Not</b> exposed to infestation after cleaning or other prescribed treatment</li> </ul>	EXEMPT
	<b>Not</b> in approved containers or have been exposed soil or to infestation after cleaning or prescribed handling	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Hay	<ul style="list-style-type: none"> <li>◆ Moved in approved containers</li> <li>◆ <b>Not</b> exposed to infestation after cleaning or other prescribed handling</li> </ul>	EXEMPT unless notified otherwise by inspector
	<b>Not</b> in approved containers or has been exposed to infestation after prescribed handling	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov

**TABLE 3-1-1 Determine if Certificate or Permit Is Required for Regulated Articles (continued)**

<b>If the article is:</b>	<b>And:</b>	<b>Then:</b>
Humus, separate or mixed with other things	Certified free from golden nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Irish potatoes (for seed)	Certified free from Golden Nematode and <b>not</b> for shipment to Puerto Rico	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	From noninfested fields for shipment to Puerto Rico and certified free from Golden Nematode	1. May SHIP in new burlap bags 2. ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov

**TABLE 3-1-1 Determine if Certificate or Permit Is Required for Regulated Articles (continued)**

If the article is:	And:	Then:
Irish potatoes ( <b>not</b> for seed)	<ul style="list-style-type: none"> <li>◆ Graded at an approved grader or washed free of soil</li> <li>◆ Was <b>not</b> exposed to infestation after cleaning or other prescribed handling and packaged in approved containers</li> </ul>	EXEMPT from regulation
	<b>Not</b> graded at an approved grader or not washed free of soil or exposed to infestation after cleaning or other prescribed handling or <b>not</b> packaged in approved containers	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
	Is from noninfested fields for shipment to Puerto Rico	<ol style="list-style-type: none"> <li>1. May SHIP in new burlap bags</li> <li>2. ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine</li> </ol>
	Is from noninfested fields for shipment to Puerto Rico but <b>lacks</b> the required PPQ Form 540	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.J.Kepich@aphis.usda.gov
Manure, decomposed; and separate or mixed with other things	Certified free from golden nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov

**TABLE 3-1-1 Determine if Certificate or Permit Is Required for Regulated Articles (continued)**

<b>If the article is:</b>	<b>And:</b>	<b>Then:</b>
Muck	Certified free from golden nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Peat	Certified free from golden nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov

**TABLE 3-1-1 Determine if Certificate or Permit Is Required for Regulated Articles (continued)**

If the article is:	And:	Then:
Plant crowns and roots for propagation	Certified free from golden nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Plant litter of any kind	<ul style="list-style-type: none"> <li>◆ Moved in approved containers</li> <li>◆ <b>Not</b> exposed to infestation after cleaning or other prescribed handling</li> </ul>	EXEMPT from regulation unless otherwise notified by inspector
	<b>Not</b> in approved containers or has been exposed to infestation after cleaning or other prescribed handling	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Plants with roots	Certified free from golden nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov

**TABLE 3-1-1 Determine if Certificate or Permit Is Required for Regulated Articles (continued)**

<b>If the article is:</b>	<b>And:</b>	<b>Then:</b>
Plants, aquatic with roots, and contain soil or soil is attached to roots	Certified free from golden nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
	Soil-free ( <b>does not</b> contain soil; <b>no</b> soil is attached to roots)	EXEMPT from regulation unless notified otherwise by inspector; EXIT this manual
Root crops (other than Irish potatoes and sugar beets)	<ul style="list-style-type: none"> <li>◆ <b>Not</b> exposed to infestation after cleaning or other prescribed handling</li> <li>◆ Moved in approved containers</li> </ul>	EXEMPT from regulation unless notified otherwise by inspector; EXIT this manual
	<b>Not</b> in approved containers or have been exposed to infestation after cleaning or other prescribed handling	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Soil, separate or mixed with other things	Certified free from golden nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Soil samples, for shipment to an approved laboratory	Shipped to an approved laboratory	EXEMPT from regulation; certificate or permit is <b>not</b> required

**TABLE 3-1-1 Determine if Certificate or Permit Is Required for Regulated Articles (continued)**

If the article is:	And:	Then:
Soil samples, shipment to <b>other than</b> an approved laboratory	Certified free from golden nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Soybeans	<ul style="list-style-type: none"> <li>◆ Harvested in bulk or directly into approved containers</li> <li>◆ <b>Not</b> exposed to infestation after cleaning or other prescribed handling</li> <li>◆ Soybeans and containers thereof have <b>not</b> come into contact with the soil</li> </ul>	EXEMPT unless notified otherwise by an inspector; EXIT this manual
	<b>Not</b> harvested in bulk or <b>not</b> harvested directly into approved containers or have been exposed to infestation after cleaning or other prescribed handling or soybeans or containers have come into contact with the soil	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Straw	<ul style="list-style-type: none"> <li>◆ Moved in approved containers</li> <li>◆ <b>Not</b> exposed to infestation after cleaning or other prescribed handling</li> </ul>	EXEMPT unless notified otherwise by inspector
	<b>Not</b> in approved containers or straw or containers have been exposed to infestation after cleaning or other prescribed handling	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Sugar beets		<ol style="list-style-type: none"> <li>1. <b>DO NOT MOVE</b> (see <b>Sugar Beets on page 3-1-22</b>)</li> <li>2. CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov</li> </ol>
<b>Other than</b> listed above		GO to <b>Table 3-1-2, "Determine if Certificate or Permit Is Required for Used Containers and Used Equipment," on page 3-1-17</b>

## Approved Containers and Equipment

### Containers and Vehicles Approved for Use Without Certification

Only the following types of containers are approved to be used for the purposes described in this manual:

- ◆ New paper bags
- ◆ New consumer packages of most material
  - ❖ **Cannot** be cloth
  - ❖ **Cannot** be burlap unless used for export shipment and approved by the importing country



Important

All new burlap bags to be used for export **only** and approved by the importing country **must** be kept in storage in the U.S. prior to use and **must** be clearly marked and labeled "For Export".

If free of soil and approved by an inspector, then the following containers may also be used to ship the regulated articles listed in this manual:

- ◆ Boxcars
- ◆ Crates
- ◆ Pallet boxes
- ◆ Trucks

### Used Containers and Other Used Equipment

Certain other used containers and used equipment are regulated, but **must** be proper cleaning, disinfecting, and certification as required before use. See [Table 3-1-2](#) below. See [Used Farm Equipment, Mechanized Soil Moving Equipment, Used Containers, Used Farm Tools, and Other Similar Articles](#) on page 3-1-25 for cleaning and disinfection instructions.

**TABLE 3-1-2 Determine if Certificate or Permit Is Required for Used Containers and Used Equipment**

<b>If the item is:</b>	<b>And:</b>	<b>Then:</b>
Containers: ◆ Burlap bags ◆ Boxes ◆ Crates ◆ Other used farm product containers	Certified free from golden nematode	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
	<b>Not</b> free of soil or exposed to infestation	GO to <b><i>Used Farm Equipment, Mechanized Soil Moving Equipment, Used Containers, Used Farm Tools, and Other Similar Articles on page 3-1-25</i></b>
	Approved (by the appropriate State regulatory official or designee) for movement within the quarantined State, as a noncertified, regulated article, to a specific destination for processing and treatment	ATTACH PPQ Form 530, Limited Permit
	Approved (by PPQ Deputy Administrator or designee in concurrence with receiving State officials) for movement to a State outside the quarantined State, as a noncertified, regulated article, to a specific location for processing and treatment	
	<b>Lacks</b> the required PPQ Form 540 or PPQ Form 530, Limited Permit	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or daniel.j.kepich@aphis.usda.gov
Used farm tools	Cleaned free of soil and <b>not</b> exposed to infestation after cleaning or other prescribed handling	RELEASE; exempt from regulation
	<b>Not</b> cleaned free of soil or exposed to infestation after cleaning	1. CLEAN free of soil (see <b><i>Used Farm Equipment, Mechanized Soil Moving Equipment, Used Containers, Used Farm Tools, and Other Similar Articles on page 3-1-25</i></b> ) 2. VERIFY items have been cleaned and disinfected 3. RELEASE

**TABLE 3-1-2 Determine if Certificate or Permit Is Required for Used Containers and Used Equipment (continued)**

<b>If the item is:</b>	<b>And:</b>	<b>Then:</b>
◆ Used mechanized cultivating equipment	Cleaned free of soil and not exposed to infestation after cleaning or other prescribed handling	ATTACH the required PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantine
◆ Used mechanized harvesting equipment	<b>Not</b> cleaned free of soil or has been exposed to infestation after cleaning	<ol style="list-style-type: none"> <li>1. CLEAN free of soil (see <a href="#">Used Farm Equipment, Mechanized Soil Moving Equipment, Used Containers, Used Farm Tools, and Other Similar Articles on page 3-1-25</a>)</li> <li>2. VERIFY items have been cleaned and disinfected</li> <li>3. RELEASE</li> </ol>
◆ Used mechanized soil-moving equipment		
<b>Other than</b> a used article, product, or means of conveyance listed above or in <a href="#">Table 3-1-1 on page 3-1-7</a>	Inspector determined the item presents a hazard of spreading golden nematode	CONTACT the Golden Nematode Program Director, Daniel Kepich, 607-566-2846, xt 2212, or <a href="mailto:daniel.j.kepich@aphis.usda.gov">daniel.j.kepich@aphis.usda.gov</a>
	<b>Not</b> hazardous to the spread of golden nematode as determined by inspector	Exempt from regulation

## Sanitation and Treatment

Officers **must** follow the instructions in this manual as a basis for treatment or other procedures to be used in authorizing the movement of regulated articles. This manual serves as a basis for explaining such procedures to persons interested in moving articles affected by quarantine regulations.



**Only** the treatment procedures authorized in this manual may be utilized **without** special permission from the PPQ Deputy Administrator.

See the *PPQ Treatment Manual* for general instructions about regulatory fumigation. Where necessary to guide proper treatment, specifications for authorized fumigants are included.

Officers will furnish complete information to anyone interested in moving regulated articles. Officers may aid shippers in the selection of authorized procedures. The shipper may select the procedure which appears to be most practical from the shipper's standpoint.

### Sanitation

Various sanitation procedures **must** be followed from the time a field is identified as infested with golden nematode until the field is officially surveyed and confirmed negative for golden nematode to prevent the spread of the pest. Procedures deemed appropriate by the officer will be implemented following each authorized activity on an infested field. See

### Authorized Pesticides

Steam heat treatment is the established method of fumigating regulated articles. See T.406C & T.406D in the *PPQ Treatment Manual*. Methyl bromide is authorized for treatment of regulated articles for golden nematode. See [Figure 3-1-1](#).

Dosage	Temperature and Exposure Time	Reading
15 lb. per 1,000 square feet (ft.)	24 hours (hr.) @ 15.5° C (60° F) or above	180 grams (g) ounce (oz.) minimum concentration reading at 1/2 hour 120 grams (g) ounce (oz.) minimum concentration reading at 24 hours
128 (g/m <sup>3</sup> ) (8 lb./1000 ft. <sup>3</sup> ) (grams/m <sup>3</sup> = oz./1000 ft. <sup>3</sup> )	48 hours (hr.) @ 15.5° C (60° F) or above	100 g oz. minimum concentration reading at 1/2 hour 75 g oz. minimum concentration reading at 24 hours 50 g oz. minimum concentration reading at 48 hours

**FIGURE 3-1-1 Methyl Bromide Fumigation at Normal Atmospheric Pressure (NAP)**

### Certification Period

Once the infested field is treated, the certification period lasts as long the treated area is protected from recontamination.

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## Approved Treatments

### Irish Potatoes, Grade A or Grade B (Except Seed Potatoes)

Potatoes must be washed, brushed, or flumed to remove soil.

When Grade A or Grade B Irish Potatoes (except seed potatoes) meet the requirements below, then the potatoes may move from golden nematode-regulated areas **without** certification (**except not** from infested fields).

1. Fields that have received two years of a resistant variety treatment followed by a negative post-treatment survey before they are replanted to potatoes.
2. Subsequent production from such fields would move as described below:
  - A. Potatoes will be free of soil, including soil clods, soil clumps, soil peds, aggregates, etc.
  - B. Shipping container will be free of soil.
3. Subsequent potato production from formerly-infested fields which have undergone successful resistant-variety treatment verified by intensive negative survey as described in xx, may move in accordance with procedures described above.
4. Resistant varieties grown as a control treatment on infested fields **must** be graded and cleaned under the observation of an officer who will permit the movement of these potatoes when cleaned to the officer's satisfaction, and transported for consumption in approved containers (see [Approved Containers and Equipment](#) on page 3-1-16). In some instances, satisfactory cleaning may require washing, grading, and/or fluming.

### Movement of Potatoes Under Limited Permit (Except Seed Potatoes)

Potatoes (except seed potatoes) may be moved under limited permit **only** as listed below.

#### Potatoes from Noninfested Fields

If one or more of the criteria described under [Approved Treatments](#) are **not** met, then the potatoes from noninfested fields would move **only** under limited permit to an approved processing plant or marketing site.

#### Potatoes from Fields Found Infested After Planting

Potatoes from fields that are found infested after planting **must** be washed under direct supervision of an officer, and moved to an approved destination in approved containers, under limited permit.

### Ear Corn With Shucks Attached

If ear corn with shucks attached are harvested in bulk or directly into approved containers and neither the corn nor containers have come into contact with the soil, then the corn is eligible for movement **without** a certificate or permit (PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantines or PPQ Form 530, Limited Permit).

### Hay, Straw, and Plant Litter

Hay, straw, and plant litter movement shall be in approved containers in accordance with the conditions listed below:

- ◆ If free of soil, then hay, straw or plant litter is eligible for movement **without** a certificate or permit (PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantines or PPQ Form 530, Limited Permit)
- ◆ If such products are felt by a PPQ Officer to constitute a risk of moving golden nematode and the owner of the premise has been notified, then the PPQ Officer may do **either** of the following:
  - ❖ Require the product to be fumigated and a certificate to be attached (PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantines)
  - ❖ Require the product to be moved under limited permit to an approved destination (PPQ Form 530, Limited Permit)

### Plants With Roots, Bulbs, Corms, Rhizomes, Ornamental Plant Tubers, and Sod (Other than Tomato and Eggplant Transplants)

Movement of plants with roots (**other than** and **not** including tomato and eggplant transplants), bulbs, corms, rhizomes, ornamental plant tubers, and sod shall be in accordance with the conditions prescribed below:

- ◆ If from noninfested fields in regulated areas, then move under PPQ Form 540, Certificate of Federal/State Domestic Quarantines or PPQ Form 530, Limited Permit (see [Table 3-1-1 on page 3-1-7](#))
- ◆ If from infested fields or exposed fields, then **must** move under PPQ Form 530, Limited Permit, in accordance with the stipulations of the compliance agreement (see [PPQ Form 519, Compliance Agreement on page A-1-29](#)) to an approved local nonagricultural destination

### Root Crops (Other than Irish Potatoes and Sugar Beets)

Movement of root crops (other than Irish potatoes and sugar beets) shall be in approved containers in accordance with the conditions prescribed below:

- ◆ If from noninfested fields in regulated areas, then eligible for movement **without** a certificate or permit
- ◆ If from infested or exposed fields and a PPQ Officer judges such products constitute a risk of moving golden nematode and the owner of the premise has been notified in writing, then the PPQ Officer may do either of the following:
  - ❖ Require the product to be washed free of soil and the certificate (PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantines) **must** be attached
  - ❖ Require the product to be moved under limited permit to an approved destination (PPQ Form 530, Limited Permit)

### **Sugar Beets**

There is **no** known method of certifying sugar beets for movement from regulated areas. Sugar beets are prohibited from movement.

### **Small Grains**

Small grains are eligible for movement under either of the following conditions:

- ◆ If harvested in bulk or directly into approved containers and **neither** the small grains **nor** containers have come into contact with the soil, then the small grains and containers are eligible for movement **without** a certificate
- ◆ If cleaned to meet State seed sales requirements, then small grains may be moved in new burlap bags **without** a certificate or permit

### **Soybeans (Other than for Seed)**

When soybeans (other than for seed) are harvested in bulk or directly into approved containers and **neither** the soybeans **nor** the containers have come into contact with the soil, then soybeans are eligible for movement **without** a certificate or permit (PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantines or PPQ Form 530, Limited Permit).

### **Soil**

Soil for fumigation should be friable and moist but **not** wet. Lids should be removed from small boxes containing soil, and individual sacks should be opened and then placed in the fumigation chamber.

Level soil mass of bulk soil to a maximum depth of 30 centimeters (1 ft.). A minimum headspace of 15 centimeters (1/2 ft.) **must** be provided.

### **Limitations of Soil for Fumigation**

**Soil for Movement to Approved Non-agricultural Areas**

Soil moving to approved non-agricultural areas (such as urban areas), is eligible for movement under PPQ Form 530, Limited Permit, in accordance with the stipulations of a compliance agreement (see **PPQ Form 519, Compliance Agreement** on page A-1-29).

**Soil Samples, Bulk Soil, Potting Soil, and Bench Soil**

After fumigation with methyl bromide, then soil samples, bulk soil, potting soil, and bench soil may be moved with PPQ Form 540, Certificate of Federal/State Domestic Quarantines, attached. See **Steam heat treatment is the established method of fumigating regulated articles. See T.406C & T.406D in the PPQ Treatment Manual. Methyl bromide is authorized for treatment of regulated articles for golden nematode. See Figure 3-1-1.** on page 3-1-19 and **Limitations of Soil for Fumigation**, and **PPQ Form 540, Certificate of Federal/State Domestic Quarantines** on page A-1-37.



Plants intended for use as feed or food crops (except tomatoes and strawberries) can **not** be grown to maturity in soil that was fumigated with methyl bromide.

**Dry Heat Fumigation of Soil Samples**

Exposure time begins after the entire soil mass has been brought to the required temperature (see **Figure 3-1-2**).

Temperature	Exposure Time
105° C (221° F)	60 minutes
114° C (237° F)	45 minutes

**FIGURE 3-1-2 Temperature for Dry Heat Fumigation of Soil Samples**

## Steam Fumigation for Bench and Potting Soil



**Only** soil from sources that are **not** been known to be infested or exposed may be approved.

If the source of the bench or potting soil has been approved in advance by a PPQ Officer, and the bench soil or potting soil is treated in accordance with the time and temperature shown in [Figure 3-1-3](#), then the bench or potting soil may be certified.

Type of Enclosure	Temperature	Exposure Time
Greenhouse benches or other containers	82.2° C (180° F)	1 hour <sup>1</sup>

**FIGURE 3-1-3 Temperature and Exposure for Steam Fumigation of Bench and Potting Soil**

- 1 Exposure time begins when the entire soil mass reaches 82.2° C (180° F) and this temperature **must** be maintained throughout the entire treatment.

### Soil for Movement to Approved Laboratory

Soil moving to an approved laboratory **does not** require a certificate or permit.

### Tomato and Eggplant Transplants

If tomato and eggplant transplants are from nonexposed fields in a regulated area, are washed free of soil as directed by a PPQ Officer, and are packaged in approved containers with a certificate (PPQ Form 540) attached, then the transplants are eligible for movement to any destination.

There are **no** approved treatment procedures for tomato or eggplant transplants are from **infested** fields (treated or **not** treated), and no approved treatment procedures for exposed fields.

### Transplants from Greenhouse Establishments on Noninfested Property

Tomato and eggplant transplants may be produced and shipped with soil from greenhouse establishments on noninfested property, when as prescribed below:

- ◆ Source of the soil has been approved in advance by a PPQ Officer
- ◆ Approved soil has been treated with schedules as outlined in this manual and **only** soil from sources that have **not** been known to be infested or exposed to golden nematode may be approved
- ◆ Required certificates **must** be attached for each shipment (see [Table 3-1-1 on page 3-1-7](#))

If grown in a soil-free medium, then tomato and eggplant transplants may also be moved; certificates **must** be attached for each shipment.

### Used Farm Equipment, Mechanized Soil Moving Equipment, Used Containers, Used Farm Tools, and Other Similar Articles

Movement of used farm equipment, mechanized soil moving equipment, used containers, and other similar articles from the regulated area shall be in accordance with the conditions described below.

#### Noncropland and Nonhost Cropland

If the used farm equipment, used farm tools, used mechanized soil moving equipment, used containers and other similar articles have been used on noncropland and nonhost cropland, then the equipment and articles are eligible for movement **without** treatment or certification. Equipment and articles are to be inspected and certified **only** when deemed necessary by a PPQ Officer.

#### Host Cropland

If all soil can be removed by cleaning the used farm equipment, used mechanized soil moving equipment, used farm tools, used containers, and other similar articles, then clean and certify using water or steam treatment; otherwise, fumigate and certificate (PPQ Form 540, Certificate of Federal/State Domestic Plant Quarantines) equipment and articles listed above and used on golden nematode host cropland, as follows:

- ◆ If treated using water-under-pressure, then use a single orifice nozzle and all soil **must** be removed
- ◆ If treated using steam (portable steam jennies or other steam equipment), then the steam **must** remove all soil or other debris



Steam may remove loose paint on equipment and is **not** recommended for use on machinery with conveyer belts or rubber parts.

All soil and debris **must** be removed. If in the judgment of the PPQ Officer equipment and articles **cannot** be adequately cleaned by water-under-pressure (washing) or steam cleaning, then the equipment **must** be fumigated. Soil should be removed prior to fumigation. Particular attention **must** be paid to removing compacted soil. See [Figure 3-1-4](#) for fumigation dosage.

Dosage	Temperature	Reading
240 g/m <sup>3</sup> (15 lb./1000 ft. <sup>3</sup> )	24 hours @ 15.5° C (60° F) or above	180 g oz. minimum concentration reading at 1/2 hour 120 g oz. minimum concentration reading at 24 hours
128 g/m <sup>3</sup> (8 lb./1000 ft. <sup>3</sup> )	48 hours @ 15.5° C (60° F) or above	100 g oz. minimum concentration reading @ 1/2 hour 75 g oz. minimum concentration reading @ 24 hours 50 g oz. minimum concentration reading @ 48 hours

**FIGURE 3-1-4 Dosage, Temperature, and Reading for Methyl Bromide Fumigation at Normal Atmospheric Pressure (NAP), Chamber or Tarpaulin**

### **Certification Period**

Once fumigated, the certification period lasts as long the treated area is protected from recontamination.

### **Movement from Infested or Exposed Portions of Regulated Area**

When the equipment and articles are cleaned to the satisfaction of a PPQ Officer and the required PPQ Form 530, Certificate of Federal/State Domestic Quarantines, is attached to the equipment and articles, then movement of the equipment from infested or exposed fields to nonexposed portions of the regulated area.

---

## **Inspection of Grader Stations**

All grader stations under compliance agreement will be inspected to insure that stipulations of the compliance agreement are being followed on a schedule determined by the appropriate officer-in-charge. See [PPQ Form 519, Compliance Agreement](#) on page A-1-29.

## Guidelines for Releasing Land from Regulation

Land previously found infested with golden nematode which has **not** been fumigated under PPQ supervision may be released from regulations<sup>1</sup> if the land meets the criteria of one of the categories listed below.

### Category 1 Non-agricultural Land (20-year Status)

Category 1 non-agricultural land, is land which was infested with golden nematode and which has been in non-agricultural status for 20 years. This land will be released from regulation upon a review of the records to determine that the land has been in non-agricultural status for the past 20 years.

Non-agricultural land includes the following:

- ◆ Highways
- ◆ Industrial areas
- ◆ Recreational land (such as golf courses, racetracks, riding academies, etc.)
- ◆ Residential areas (including home gardens)

### Category 2 Non-agricultural Land (Less than 20-year Status)

Category 2 non-agricultural land, is land which was infested with golden nematode and which has been in non-agricultural status for less than 20 years may be released if construction for non-agricultural purposes has rendered the acreage nontillable.

Nontillable land acreage includes the following:

- ◆ Office buildings and parking complex
- ◆ Mall and parking complex
- ◆ Shopping center and parking complex

### Category 3 Agricultural Land

Category 3 agricultural land, is land which has been planted to nonhost crops. This land would be released from regulations after being planted in nonhost crops for 20 years, followed by a negative soil survey on either a 4 x 4 sampling pattern or a mechanical sampler with a level of detection of 100,000 cysts per acre in the top 4 inches of soil.

Agricultural land planted to nonhost crops includes the following:

- ◆ Fallow fields

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<sup>1</sup> Prior to 1959, infested land was removed from agricultural use because fumigation was **not** available.

- ◆ Forage crops
- ◆ Grain fields
- ◆ Nurseries
- ◆ Sod farms
- ◆ Truck farms

This land is sampled because of the close association with and possible contamination by erosion, equipment, water, wind, etc.

---

### **Seed Potato Certification for Interstate Movement**

Certification is not required for seed potatoes. Survey/soil sampling of all seed potato land is done only to confirm that the soil is free of golden nematode in New York. Work with the State of New York for all growers that have seed potatoes.

# 4

Golden Nematode  
Program Manual

## Procedures

### *Control*

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#### Contents

Introduction	page 4-1-1
Non-chemical Control	page 4-1-1
Systematic Use of Resistant Potato Varieties	page 4-1-1
Restrictions on Infested Property	page 4-1-4
Crop Management Sequence	page 4-1-4
Steam Treatment	page 4-1-5
Ro2	page 4-1-5
Chemical Control	page 4-1-5

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#### Introduction

The *Procedures* section of the *Control* chapter provides information about non-chemical methods used to treat and control the spread of golden nematode and help to eradicate golden nematode in infested areas.

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#### Non-chemical Control

The following methods of non-chemical control are used to help prevent the spread of golden nematode:

- ◆ Systematic use of resistant-varieties of potatoes to reduce golden nematode populations below detectable levels
  - ❖ Require growers are to plant resistant varieties on land treated since 1972
  - ❖ Require growers to plant resistant varieties on exposed land
- ◆ Steam heat treatment (of equipment)
- ◆ Survey
- ◆ Federal and State regulations

##### **Systematic Use of Resistant Potato Varieties**

The primary authorized control mechanism is the mandated planting of approved golden nematode resistant varieties of potatoes. The use of host-plant resistance to reduce the population of golden nematode is a biological means of pest control. Golden nematode cysts hatch when stimulated by Solanaceae potato root exudates. The nematodes then penetrate the host plant roots and establish a feeding site.

With resistant potato varieties, the juvenile nematodes exist on the roots, in part because the nematode **must** feed on live cells in the potato plant's roots, and in the resistant roots, the cells around the nematode's feeding site die, and most of the nematodes die, too. Of the few surviving nematodes, reproduction is diminished and their offspring have a lowered rate of infestation.

A list of potato varieties that are currently resistant to golden nematode are shown in [Figure 4-1-1](#).

Year Introduced	Potato Variety	Agency
1966	Peconic	Cornell University
1967	Wauseon	USDA-MD
1972	Hudson	Cornell University
1976	Atlantic	USDA-MD
1977	Campbell	Cornell University
1978	Campbell 13	Cornell University
1979	Belchip	USDA-MD <sup>1</sup>
1980	Highlat Russet	USDA-ARS <sup>2</sup>
1981	Rosa	Cornell University
1982	Simcoe	Agriculture Canada
1984	Islander	University of Maine
1985	Elba	Cornell University
1985	Hampton	Cornell University
1985	Sunrise	University of Maine
1986	Donna	Agriculture Canada
1989	Kanona	Cornell University
1989	NemaRus	USDA-MD
1990	Alleghany	Cornell University
1990	Steuben	Cornell University
1991	Castile	Cornell University and USDA-MD
1991	LaBelle	Louisiana State University
1992	Coastal Chip	USDA
1992	Sparton Pearl	Michigan State University
1992	Michigold	Michigan State University
1993	Genesee (NY78)	Cornell University
1993	St. John's (AF838-5)	University of Maine
1993	Sunchip	USDA-MD
1995	Pike	Cornell University
1995	Andover	Cornell University
1997	Salem	Cornell University
1999	Keuka Gold	Cornell University
1999	Eva	Cornell University
1999	Amey	USDA-MD
2003	Marcy	Cornell University
2003	Sante	GN Ro1 and Ro2 resistant variety Germicopa, France
2004	Fabula	HZPC, Netherlands

**FIGURE 4-1-1 List of Golden Nematode Resistant Potato Varieties**

- 1 USDA in Beltsville, Maryland.
- 2 USDA-ARS in Palmer, Alaska.

### Restrictions on Infested Property

When property is found to be infested with the golden nematode disease, the owner or operator **must** enter into an agreement with the New York State Department of Agriculture and Markets. The agreement restricts the crops that may be grown to either varieties of potatoes that are resistant to golden nematode or an approved non-host crop. A non-host crop is defined as any crop which is **not** in the *Solanaceae* family. (Potato, tomato, and eggplant are in the *Solanaceae* family).

**EXAMPLE** Examples of non-host crops are: alfalfa, carrots, corn, cucumbers, pumpkins, rye, and wheat.

### Post-resistant Variety Treatment (PRVT)

If the land is to continue in potato production, a minimum of 2 successive crop years of golden nematode-resistant potato variety **must** be grown. Following harvest of the second crop, the field will be intensively surveyed in accordance with procedures described in **Post Resistant-Variety Treatment Survey on page 2-3-13**. All surveys **must** be negative for viability. If the survey is negative, then the farmer may continue to grow either golden nematode-resistant varieties or non-host crops.

The farmer may also enter into an approved pest management program agreement. See **Pest Management Program**.

### Crop Management Sequence

### Pest Management Program

Farmers or growers may enter an approved pest management program that uses resistant varieties, non-host, and susceptible varieties in a 4-year crop rotation. See

Following two consecutive crop years of resistant varieties grown on infested land and a negative survey, growers may enter the crop rotation system at the resistant variety or non-host year.



The approved crop rotation system **cannot** be entered at the susceptible variety year under any case or circumstance.

If a grower chooses to enter the pest management program at year 3 (non-host) and a susceptible variety is planted the following year, then the field must be surveyed after the susceptible variety is harvested in accordance with the survey procedures. If this survey is negative for golden nematode, then **no** further surveys need to be conducted on this land as long as the approved rotation system is followed.

An additional option is available for growers who wish to plant a non-host crop on an infested field without having to plant a resistant potato variety for 2 years. In this option, a grower applies a single application of a registered fumigant at the legal dosage rate to the regulated property. The planting of potatoes or other host crops on this field is still **prohibited**.

### **Steam Treatment**

Refer to the *PPQ Treatment Manual* for detailed steam heat treatment instructions.

### **Ro2**

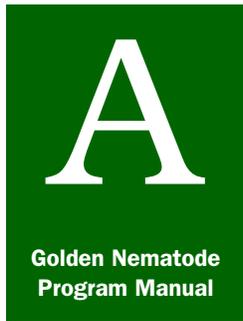
Special crop rotation procedures are required on land where Ro2 has been detected. Grower options are very limited because Ro2 resistant potato varieties are still being developed. At present, the Sante potato variety is resistant to both Ro1 and Ro2 golden nematode.

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## **Chemical Control**

Chemical control procedures are **no** longer approved for routine program use in the State of New York.





# Appendix A

## *Forms and Worksheets*

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### Contents

Introduction	<b>page A-1-2</b>
Golden Nematode Laboratory Sample Processing Daily	<b>page A-1-3</b>
Purpose	<b>page A-1-4</b>
Instructions	<b>page A-1-4</b>
Distribution	<b>page A-1-6</b>
Golden Nematode Farm Survey Questionnaire	<b>page A-1-7</b>
Purpose	<b>page A-1-8</b>
Instructions	<b>page A-1-8</b>
Distribution	<b>page A-1-8</b>
Golden Nematode Survey Data Worksheet	<b>page A-1-9</b>
Purpose	<b>page A-1-11</b>
Instructions	<b>page A-1-11</b>
Distribution	<b>page A-1-12</b>
Sample Storage Sheet	<b>page A-1-13</b>
Purpose	<b>page A-1-14</b>
Instructions	<b>page A-1-14</b>
Distribution	<b>page A-1-14</b>
Weekly Summary Record	<b>page A-1-15</b>
Purpose	<b>page A-1-16</b>
Instructions	<b>page A-1-16</b>
Distribution	<b>page A-1-16</b>
PPQ Form 312, Golden Nematode Survey	<b>page A-1-17</b>
Purpose	<b>page A-1-21</b>
Instructions	<b>page A-1-21</b>
Distribution	<b>page A-1-23</b>
PPQ Form 333, Cyst Nematode Field Survey Log	<b>page A-1-24</b>
Purpose	<b>page A-1-25</b>
Instructions	<b>page A-1-25</b>
Distribution	<b>page A-1-25</b>
PPQ Form 391, Specimens for Determination	<b>page A-1-26</b>
Purpose	<b>page A-1-27</b>
Instructions	<b>page A-1-27</b>
Distribution	<b>page A-1-28</b>
PPQ Form 519, Compliance Agreement	<b>page A-1-29</b>
Purpose	<b>page A-1-33</b>
Instructions	<b>page A-1-33</b>
Distribution	<b>page A-1-34</b>
PPQ Form 530, Limited Permit	<b>page A-1-35</b>
Purpose	<b>page A-1-36</b>
Instructions	<b>page A-1-36</b>
Distribution	<b>page A-1-36</b>
PPQ Form 540, Certificate of Federal/State Domestic Quarantines	<b>page A-1-37</b>
Purpose	<b>page A-1-38</b>
Instructions	<b>page A-1-38</b>
Distribution	<b>page A-1-38</b>

## **Introduction**

The *Forms* appendix provides examples of Golden Nematode program logs and worksheets; PPQ forms, certificates, and permits; and instructions for completing and distributing the worksheets and forms.

**Golden Nematode Laboratory Sample Processing Daily**

GOLDEN NEMATODE LABORATORY SAMPLE PROCESSING DAILY					
BEAKER NO.	COLL. NO.	BAG NO.	BEAKER NO.	COLL. NO.	BAG NO.
1.			26.		
2.			27.		
3.			28.		
4.			29.		
5.			30.		
6.			31.		
7.			32.		
8.			33.		
9.			34.		
10.			35.		
11.			36.		
12.			37.		
13.			38.		
14.			39.		
15.			40.		
16.			41.		
17.			42.		
18.			43.		
19.			44.		
20.			45.		
21.			46.		
22.			47.		
23.			48.		
24.			49.		
25.			50.		

**FIGURE A-1-1 Example of Golden Nematode Laboratory Sample Processing Daily**

### **Purpose**

The *Golden Nematode Laboratory Sample Processing Daily* sheet is used by the Laboratory Leader to assign beaker numbers to sample collections, to complete other forms, to reconcile samples with a positive or negative determination, and to determine who made the determination on a particular bag or beaker. The sheet is also used by Biological Laboratory Technicians to record the beaker numbers assigned on the sheet to the sample bags in each collection before processing.

### **Instructions**

Information from the sample storage sheet is transferred to this sheet for beaker number assignment. See **Sample Storage Sheet** on page A-1-13 and **Figure 2-4-13** on page 2-4-22 Continue to **Table A-1-2** on page-A-1-5 to complete the *Golden Nematode Laboratory Sample Processing Daily* sheet.

**TABLE A-1-2 Instructions for Completing Golden Nematode Laboratory Sample Processing Daily**

Block	Completed by	Instructions
BEAKER NO.	Laboratory Leader	<p>This block column is pre-numbered 1-50 on the front side of the sheet and 51-83 on the back. Beakers are numbered 1-83 in the laboratory. These beaker numbers are used throughout sample processing to track samples</p> <ol style="list-style-type: none"> <li>1. TAKE the Sample Storage sheet from the rack room shelf</li> <li>2. SEE the Sample Storage sheet for the collection number that is ready for sample processing</li> <li>3. ENTER the collection number in the first blank BEAKER NO. block</li> </ol>
COLL. NO.	Biological Laboratory Technician	<ol style="list-style-type: none"> <li>1. If you have collected and placed all the cysts in a vial, then GO to the BEAKER NO. column</li> <li>2. LOCATE the sample bag collection number and beaker number on this form and CONFIRM the actual beaker sample you are reading and the sample bag collection number listed under BEAKER NO. match</li> <li>3. ENTER under COLL. NO. the number of cysts collected for identification</li> </ol>
BAG NO.	Laboratory Leader	<ol style="list-style-type: none"> <li>1. FIND the sample bag collection number listed in the BEAKER NO. block</li> <li>2. SEE the Sample Storage sheet and find the identical collection number (under COLLECTION) and the number of samples in the collection (under NO. OF SAMPLES)</li> <li>3. GO back to the Golden Nematode Laboratory Sample Processing Daily Sheet (Daily Sheet) in the BEAKER NO. column and LOCATE the sample bag collection number</li> <li>4. GO across the row to the BAG NO. column, ENTER a 1, and continue entering the sample bag numbers until you have listed every bag in the collection</li> </ol> <div style="border: 1px solid black; padding: 5px; margin: 10px 0;"> <p><b>EXAMPLE</b> There are 18 sample bags in the collection DEM-02. The collection number is listed in BEAKER NO. block 9., you will enter sample bag number 1 in the BAG NO. block across from BEAKER NO. 9, and enter 2 across from BEAKER NO. 10., and continue until you enter an 18 under the BAG NO. block across from BEAKER NO. 26.</p> </div> <ol style="list-style-type: none"> <li>5. If there are more than 83 bags in a collection, then start a new Daily Sheet (place a 2 in upper right corner of sheet) and continue numbering at BEAKER NO. 1 again</li> </ol>

### **Distribution**

Distribute the completed Golden Nematode Laboratory Sample Processing Daily sheet as follows:

- ◆ Biological laboratory aide clips each completed sheet beneath the blank sheets on the clipboard
- ◆ Laboratory Leader removes the completed sheets weekly and files in the golden nematode file folder



## Purpose

The Golden Nematode Farm Survey Questionnaire is used to gather information during the PPQ Plant Health Safeguarding Specialist's (or designee, PPQ Technician) pre-survey interview with the grower/farmer.

## Instructions

Complete the Golden Nematode Farm Survey Questionnaire as shown in [Table A-1-3](#). After the form is completed the PPQ PHSS will review the form with the PPQ Technician and make sure everything is in line for the upcoming survey.

**TABLE A-1-3 Instructions for Completing Golden Nematode Farm Survey Questionnaire**

Block		Instructions
1	COUNTY	ENTER the name of the county where the land for survey is located
2	YEAR	Enter the year the survey will be conducted (If a confirmation survey, then interview could take place in the fall of one year, but the survey won't be conducted until the next year)
3	GROWER	ENTER the name of the grower or the farm
4	DATE	ENTER the date the interview is being conducted
5	TOTAL ACRES IN POTATOES	ENTER the total number of acres planted to potatoes (all varieties)
6	TOTAL ACRES IN RESISTANT VARIETY POTATOES	ENTER the total number of acres planted in resistant varieties (RV) of potatoes
7	TOTAL ACRES TO SURVEY	SUBTRACT the number listed in Block 6 from the number listed in Block 5 and ENTER the difference in this block
8	SURVEY METHOD	PLACE an X in the appropriate block
	MECHANICAL	If a mechanical survey will be conducted, place an X in this block; otherwise leave blank
	MANUAL	If a manual survey will be conducted, place an X in this block; otherwise leave blank
	FIELD NO.	ENTER the field number to be surveyed
	VARIETY	ENTER the variety of potatoes planted in the field to be surveyed. If more than one variety planted, then list all varieties

## Distribution

Distribute the questionnaire as follows:

xx





## Purpose

The *Golden Nematode Survey Data Worksheet* is used to collect and record information during an interview with the grower or farmer prior to survey.

## Instructions

The is completed by the PPQ Office/Plant Health Safeguarding Specialist (or designee, PPQ Technician) during the pre-survey interview with the farmer/grower. The Plant Health Safeguarding Specialist will review and give the worksheet to the Laboratory Leader. The Laboratory Leader will then transfer the information into a spreadsheet. Complete the Golden Nematode Survey Data Worksheet as instructed in [Table A-1-4](#).

**TABLE A-1-4 Instructions for Completing Golden Nematode Survey Data Worksheet**

Block	Instructions
1 COUNTY	ENTER the name of the county where land to survey is located
2 YEAR	ENTER the year the survey will be conducted
3 GROWER	ENTER the name of the grower or the name of the farm where for the survey
4 DATE	ENTER the date the interview is being conducted to complete this questionnaire
5 TOTAL ACRES IN POTATOES	ENTER the total number of acres planted to potatoes
6 TOTAL ACRES (RESISTANT VARIETIES)	ENTER the total number of acres planted to resistant varieties of potatoes
7 TOTAL ACRES TO SURVEY	ENTER the total number of acres to survey Total acres of potatoes - Total acres/resistant varieties = Total acres to survey
FIELD NUMBER	If there is a historical record, then obtain the field number from the file and ENTER the field number (prior to survey/questionnaire interview or before survey)
NON-EXPOSED ACRES	ENTER the number of acres surveying as non-exposed (to golden nematode); if land is <b>not</b> regulated then enter the number of acres as non-exposed; if none, then leave blank
EXPOSED ACRES	ENTER the number of exposed acres; if none leave blank  <div style="display: flex; align-items: center;">  <div style="margin-left: 10px;"> <p><b>Important</b></p> <p>If a grower has acreage elsewhere that are regulated, then all the grower's acreage is counted as exposed acres.</p> </div> </div>
SEED ACRES	If any acreage is planted to seed potatoes, then ENTER the number of acres; otherwise leave blank
Ro2 ACRES	If the land or field had a previous positive find for the Ro2 golden nematode, then enter the total acres; otherwise leave blank

**TABLE A-1-4 Instructions for Completing Golden Nematode Survey Data Worksheet (continued)**

Block	Instructions		
POST RV TREATMENT ACRES	If the acreage is planted to resistant variety (RV) potatoes, then enter the number of acres (survey the first year after RV harvest); otherwise, leave blank		
1ST SUSCEPTIBLE AFTER TREATMENT ACRES	If the acreage is planted is planted to a non-resistant variety the first year after resistant variety (RV) treatment 2-3 years previous, the enter the number of acres; otherwise leave blank		
MECH 200K	If a Mechanical Survey 200K (3 wheels, 4 probes each) is recommended, then PLACE an X in this block; otherwise leave blank		
MECH 500K	If a Mechanical Survey 500K (2 wheels, 4 probes each) is recommended, then PLACE an X in this block; otherwise leave blank		
MAN 4 X 8	If a Manual Survey 4 x 8 block method is recommended, then PLACE an X in this block; otherwise leave blank		
NOTES	ENTER any other relevant information  <table border="1" data-bbox="829 919 1414 1020"> <tr> <td data-bbox="829 919 984 1020"><b>EXAMPLE</b></td> <td data-bbox="984 919 1414 1020">Field DEM-02 planted 1/3 RV potatoes; 1/3 susceptible potatoes; 1/3 corn.</td> </tr> </table>	<b>EXAMPLE</b>	Field DEM-02 planted 1/3 RV potatoes; 1/3 susceptible potatoes; 1/3 corn.
<b>EXAMPLE</b>	Field DEM-02 planted 1/3 RV potatoes; 1/3 susceptible potatoes; 1/3 corn.		

**Distribution**

Distribute the Golden Nematode Survey Data Worksheet as follows:

- ◆ Plant Health Safeguarding Officer gives the field completed copy to the Laboratory Technician
- ◆ Laboratory technician maintains both copies in the work unit file

Sample Storage Sheet

# AVOCA SAMPLE STORAGE

DATE	COLLECTION	NO OF SAMPLES	SOIL TYPE	RACK	SHELF NOS.	PROCESSED	PROCESSED	DATE	COLLECTION	NO OF SAMPLES	SOIL TYPE	RACK	SHELF NOS.
0/5	JED-3		MIN	I	2, 3, 5, 6			10/24	LRK-4	7	MIN	I	34
0/6	AMD-6	37	MIN	I	5, 6			10/24	LRK-5	5	MIN	I	34
	AMD-5		MIN	I	10			10/24	LRK-6	4	MIN	I	33
0/11	JLS-4	81	MIN	I	22, 25			10/24	LRK-7	4	MIN	I	33
0/11	AMD-7	64	MIN	I	4, 14			10/24	LRK-8	5	MIN	I	33
10/12	JLS-6	7	MIN	I	6			10/25	AMD-19	25	MIN	I	11, 12
0/12	JLS-5	40	MIN	I	10-8			10/28	MZK-22	29	MIN	J	23
10/2	AMD-8	49	MIN	I	16, 25, 26			10/28	MZK-21	15	MIN	J	23
10/2	MZK-11	36	MIN	I	17, 20			10/28	MZK-20	18	MIN	J	24
0/12	MZK-10	46	MIN	I	27			11/2	JLS-20	8	MIN	J	26
0/13	JLS-7	70	MIN	I	35-40			11/2	JLS-19	26	MIN	J	26
0/13	AMD-9	22	MIN	I	13			11/2	AMD-21	28	MIN	J	27, 2
1/13	MZK-12	45	MIN	I	18, 19			11/7	LRK-16	7	MIN	I	35
1/14	RHB-3	110	MIN	J	2-6			11/7	LRK-15	6	MIN	J	35
0/17	JLS-8	55	MIN	I	29, 30, 32			11/7	LRK-12	14	MIN	J	35
0/18	AMD-11	22	MIN	J	12								
10/18	AMD-12	17	MIN	J	13								
0/18	AMD-10	35	MIN	J	14, 15								
0/18	AMD-13	19	MIN	J	15, 16								
0/19	JLS-10	24	MIN	J	057								
0/19	JLS-9	37	MIN	J	8, 9								
10/19	AMD-15	36	MIN	J	16, 17								
10/19	AMD-14	34	MIN	J	18, 17								
10/20	AMD-18	3	MIN	I	3								
10/20	JLS-13	13	MIN	J	10								

FIGURE A-1-5 Example of Sample Storage Sheet

### Purpose

The Sample Storage Sheet provides is used to record and track a collection from the time each sample bag is brought into the Work Unit facility, and placed onto the shelves, until the collection is moved from the rack room to the sample wash room for processing.

### Instructions

The crew leader completes this form upon each's collection's arrival at the Work Unit rack room. Continue to [Table A-1-5](#).

**TABLE A-1-5 Instructions for Completing Sample Storage Sheet**

Block	Instructions
DATE	ENTER the date the collection is placed in the rack room
COLLECTION	ENTER the collection number from the top row of the bag (field number)
NO OF SAMPLES	ENTER the number of sample bags in the collection (located on the lower left of the bag; number indicated with END)
SOIL TYPE	ENTER Mineral or Muck (the type of sample soil in the collection)
RACK	ENTER the rack number where the collection is stacked
SHELF NOS.	ENTER the shelf numbers where the collection is stacked
PROCESSED	LEAVE blank

### Distribution

Place the sheet on the Rack Room clipboard. After the sheet is completely filled; all samples in every collection listed on the sheet are dried; and the information has been transferred to the Golden Nematode Laboratory Sample Processing Daily Sheet, then the Laboratory Leader will draw a line through every collection recorded to indicate the transfer of information. Once all samples listed on a completed sheet have been moved, then the form is no longer needed in the Rack Room and can be maintained in the Laboratory file or destroyed.



### Purpose

The Weekly Summary Record is used to xx.

### Instructions

The Weekly Summary Record is completed by the xx.

**TABLE A-1-6 instructions for the Weekly Summary Record**

BLOCK	Instructions
PERIOD	ENTER the first and last day of the period
METHOD	
MDN	
OPERATOR	
SAMPLES	
ACRES	
POSITIVE/NEGATIVE	
COUNTY	
DATE	

### Distribution

xx



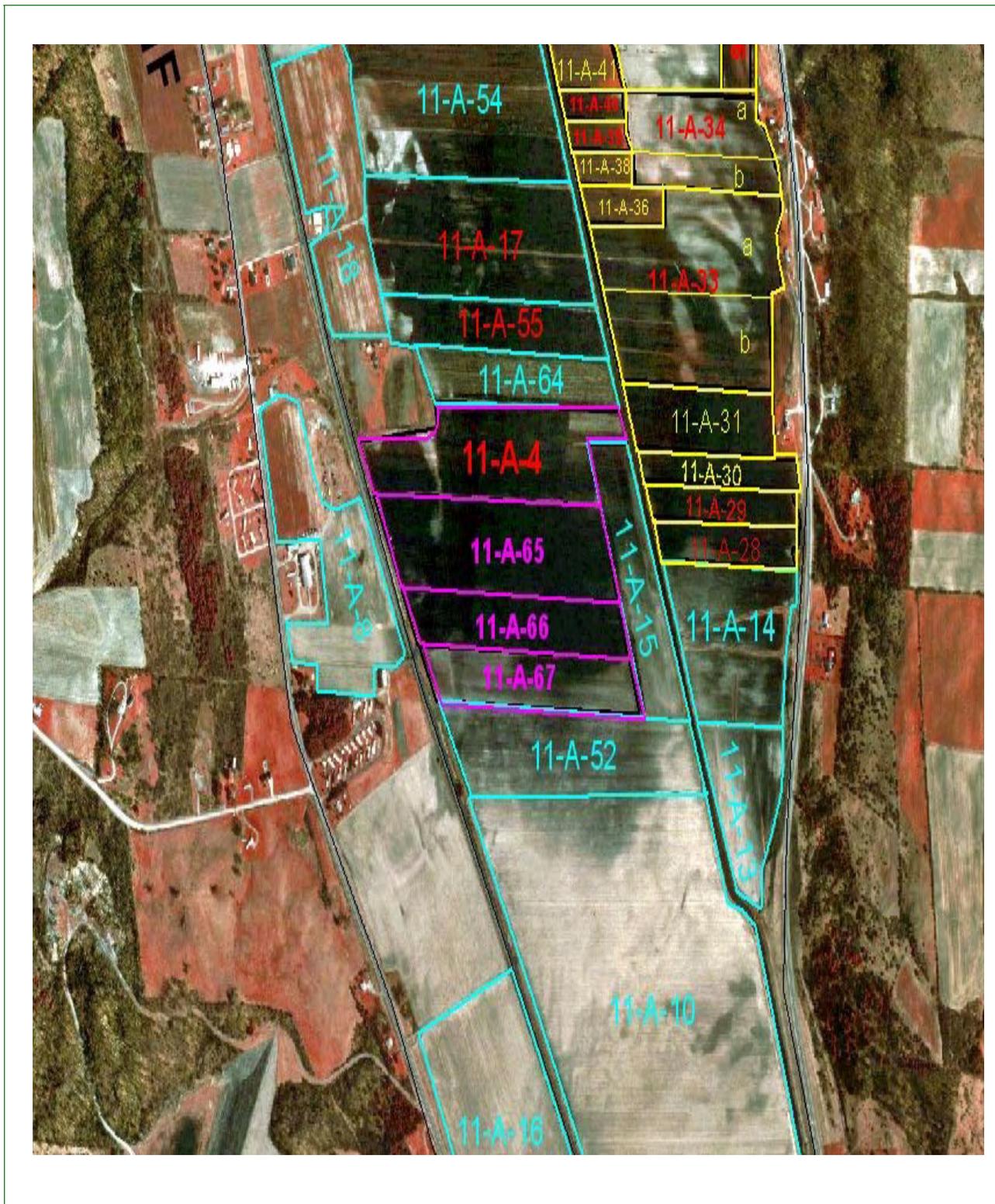


FIGURE A-1-8 Example of GIS Image of Survey Site (ArcView)



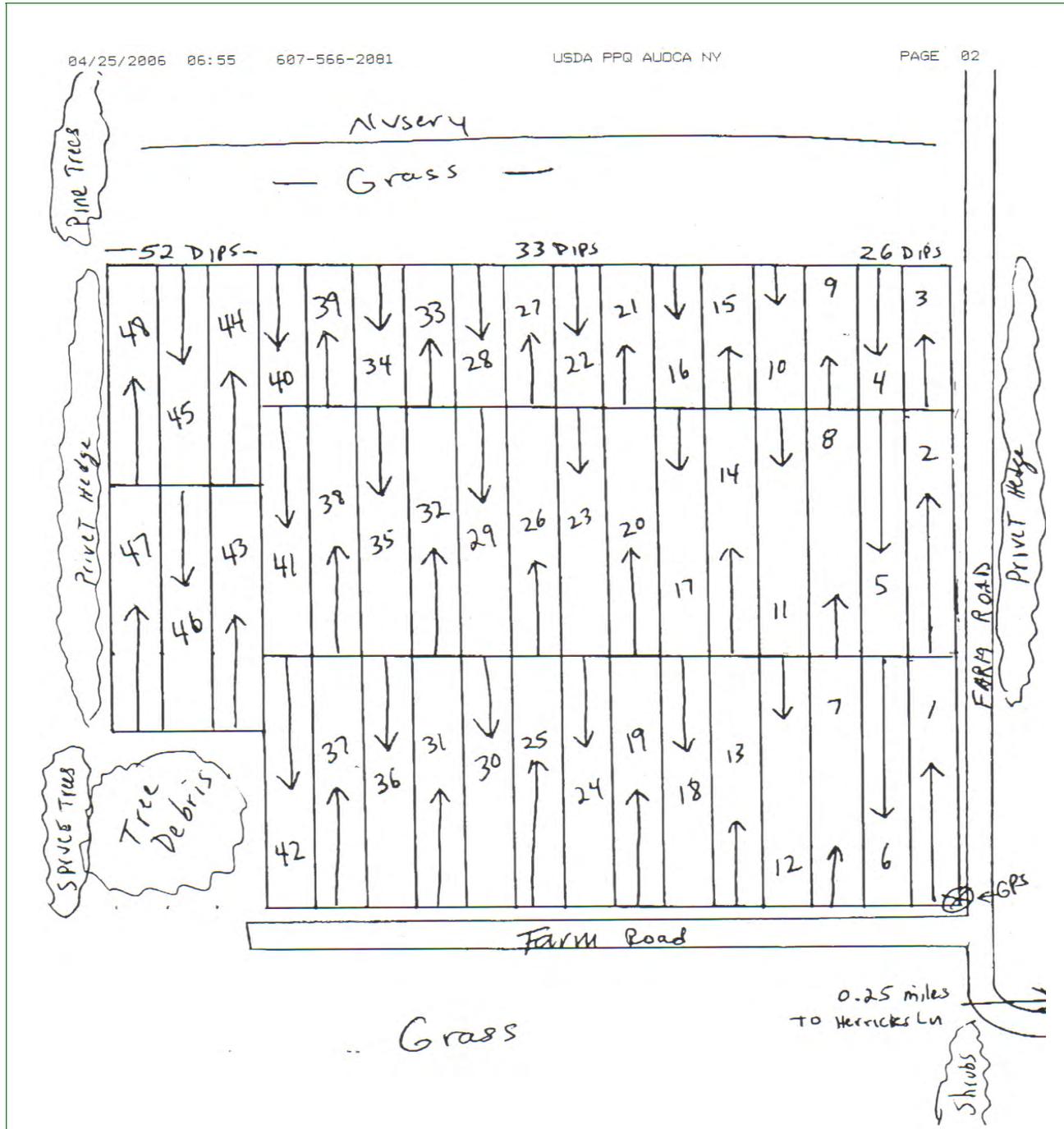


FIGURE A-1-10 Example of Hand-Drawn Nursery Survey Map

## Purpose

PPQ Form 312, *Golden Nematode Survey*, is used to document information collected during field surveys for golden nematode. A map of the property inspected is placed on the reverse side of the map. The map is important for relocating and returning to the same field in the event that golden nematode is found or further survey is needed.

## Instructions

The upper half of the front side and the reverse side of the form should be completed by the collecting officer or crew leader.

The lower half of the front side is completed by laboratory personnel. The remainder is completed by the crew leader in the field when taking the survey. The Plant Health Safeguarding Specialist will review the completed PPQ form 312.

The Plant Health Safeguarding Specialist or PPQ Technician will obtain a GIS map of the survey site (Goggle Maps Satellite, and add the GPS reading NW (latitude/longitude) on the field map. The map is then attached to the reverse side of PPQ Form 312.

If a satellite map is **not** available, the crew leader draw a simple diagram of the property inspected and indicate NW on the map. This diagram is important for relocating and returning to the same field in the event golden nematode is found or further survey is needed. The rough sketch should include enough landmarks, telephone pole numbers, and road names so that the field may be easily located. See a GIS example in [Figure A-1-8 on page A-1-18](#), and hand-drawn maps in [Figure A-1-9 on page A-1-19](#) and [Figure A-1-10 on page A-1-20](#).

During the sample survey, the crew leader will use the map to indicate the number of dips taken from first row and last row of each field sampled.

Continue to [Table A-1-9 on page-A-1-27](#) and follow the instructions for completing the form.

**TABLE A-1-7 Instructions for Completing PPQ Form 312, Golden Nematode Survey**

Block		Completed by	Instructions
1	STATE	Crew Leader	If not preprinted, then LIST the State where the collection was made
2	COUNTY	Crew Leader	LIST the county where the samples were taken
3	DATE OF SURVEY	Crew Leader	LIST the date of the survey (if more than one day, then list date first through last days)
4	COLLECTION NUMBER	PPQ PHSS	LIST the collection number
5	FIELD NUMBER	PPQ PHSS	LIST the field number (from historical files or maps in the work unit)
6	LATTITUDE/ LONGITUDE OF ENTRANCE POINT	PPQ PHSS	LIST the latitude and longitude of entrance point to the field (GPS coordinates)
7	NAME OF FARM OPERATOR		LIST the name of the farmer or grower where the collection was made
8	MAILING ADDRESS		LIST the farm operator's mailing address
9	FIELD LOCATION		ENTER a short narration of the location of the field on the property; may also list odometer readings for mileage directions
10	TYPE OF SURVEY		PLACE an X in the appropriate box to indicate the type of survey (from GN Survey Data Worksheet; MECH 200K, MECH 500K, or Manual 4 x 8)
11	NO. SAMPLES	Crew Leader	ENTER the total sample bags taken in the collection
12	SURVEY PATTERN	PPQ PHSS or Crew Leader	PLACE an X in the appropriate survey pattern box (8x8, 4x8, 4x4, 2x2)
13	METHOD OF SURVEY		PLACE an X in the box to indicate the method of survey taken (manual or mechanical (200K) or (500K))
14	NO. ACRES SURVEYED	Crew Leader	LIST the quantity of acres surveyed
15	SOIL TYPE	PPQ Officer or PPQ Tech	LIST the type of soil in the survey (mineral or muck)
16	FIELD STATUS AT TIME OF SURVEY		PLACE an X in the box to indicate the status of the field at the time of survey (plowed, cover crop, other (crop name, etc.); if planted to potatoes then ENTER "Potatoes"
17	NAMES OF COLLECTORS	Crew leader	LIST the names of the crew members collecting the samples

**TABLE A-1-7 Instructions for Completing PPQ Form 312, Golden Nematode Survey (continued)**

Block		Completed by	Instructions
18	REMARKS		LIST any remarks related to the area survey area
19	DETERMINED BY	PPQ Identifier	Leave the remainder of the blocks blank, unless an in-house identifier has the official authority to make identifications
	POSITIVE SAMPLE NUMBER	PPQ Identifier	
	NO. OF CYSTS	PPQ Identifier	
	IDENTIFICATION OF SLIDES AND VIALS FILED	PPQ Identifier	
	POSTIVE SAMPLE NO.	PPQ Identifier	
	NO. OF CYSTS	PPQ Identifier	
	IDENTIFICATION OF SLIDES AND VIALS FILED	PPQ Identifier	
19	DETERMINED BY	PPQ Identifier	
20	DATE	PPQ Identifier	

### Distribution

Distribute PPQ Form 312 as follows:

- ◆ File the original in the program folder
- ◆ If the field is confirmed as infested, then file in the infested field folder
- ◆ Send a copy to the Supervisor (xx list Western Region Supervisor's name and FAX or address and Eastern Region Supervisor's name and FAX or address)

**PPQ Form 333, Cyst Nematode Field Survey Log**

CYST NEMATODE FIELD SURVEY LOG			COUNTY Wyoming	STATE New York			
			PERIOD	NAME OF INSPECTOR TR Bourgeois			
COLLECTION NUMBER	COLL. DATE	OPERATOR	FIELD NUMBER	MECHANICAL		MANUAL	
				Samples	Acres	Samples	Acres
RHB - 4	10-21-05	MARQUART	II 2-A-11	30	13		
RHB - 9	10-24-05	McCORMICK	II 2-B-19	130	64		
RHB - 10	10-28-05	McCORMICK	II 2-A-56	58	27		
TOTAL				218	104		

FIGURE A-1-11 Example of PPQ Form 333, Cyst Nematode Field Survey Log

### Purpose

PPQ Form 333, Cyst Nematode Field Survey Log, is used by the Laboratory Leader to reconcile PPQ form 312.

### Instructions

PPQ Form 333 is completed by the crew leader to record the collections made each week.

**TABLE A-1-8 Instructions for Completing PPQ Form 333, Cyst Nematode Field Survey Log**

Block		Instructions
COUNTY		ENTER the name of the county for each field surveyed
STATE		ENTER the name of the State where the survey is conducted
PERIOD		ENTER the first and last dates of the week for the survey period
NAME OF INSPECTOR		ENTER the name of crew leader's last name
COLLECTION NUMBER		ENTER the sample collection number
COLL. DATE		ENTER the date the survey is conducted
OPERATOR		ENTER the name of the farm or the farm operator's name
FIELD NUMBER		ENTER the number of the field surveyed
MECHANICAL	Samples	If a mechanical survey was conducted, then ENTER the number of samples collected; otherwise leave blank
	Acres	If a mechanical survey was conducted, then ENTER the number of acres surveyed
MANUAL	Samples	If a manual survey was conducted, then ENTER the number of samples collected; otherwise leave blank
		If a manual survey was conducted, then ENTER the number of acres surveyed; otherwise leave blank
TOTAL		ENTER the total number of samples listed
		ENTER the total number of acres surveyed

### Distribution

The crew leader will give the completed PPQ Form 333 to the Laboratory Leader weekly. File the form in the Work Unit office.

## PPQ Form 391, Specimens for Determination

This report is authorized by law (7 U.S.C. 147a). While you are not required to respond your cooperation is needed to make an accurate record of plant pest conditions. *See reverse for additional OMB information.* **FORM APPROVED OMB NO. 0579-0010**

**U.S. DEPARTMENT OF AGRICULTURE  
 ANIMAL AND PLANT HEALTH INSPECTION SERVICE  
 SPECIMENS FOR DETERMINATION**

Instructions: Type or print information requested. Press hard and print legibly when handwritten. Item 1 - assign number for each collection beginning with year, followed by collector's initials and collector's number. Example (collector, John J. Dingle): 83-JJD-001.  
**Pest Data Section** - Complete Items 14, 15 and 16 or 19 or 20 and 21 as applicable. Complete Items 17 and 18 if a trap was used.

**FOR IIBIII USE**  
 LOT NO.  
 PRIORITY

1. COLLECTION NUMBER  
 05-ENJ-001 I-11-D-5

2. DATE  
 MO DA YR  
 01 03 05

3. SUBMITTING AGENCY  
 State Cooperator  PPQ  Other \_\_\_\_\_

4. NAME OF SENDER  
 Edward N. Jones

5. TYPE OF PROPERTY (Farm, Feedmill, Nursery, etc.)  
 Farm

6. ADDRESS OF SENDER  
 8237 Kanona Road

7. NAME AND ADDRESS OF PROPERTY OR OWNER  
 ABC Farms  
 R.D. #2

Avoca, NY  
 ZIP 14809

Arkport, NY COUNTRY/COUNTY Steuben

8. REASON FOR IDENTIFICATION ("X" ALL Applicable Items)

A.  Biological Control (Target Pest Name ) E.  Livestock, Domestic Animal Pest  
 B.  Damaging Crops/Plants F.  Possible Immigrant (Explain in REMARKS)  
 C.  Suspected Pest of Regulatory Concern (Explain in REMARKS) G.  Survey (Explain in REMARKS)  
 D.  Stored Product Pest H.  Other (Explain in REMARKS)

9. IF PROMPT OR URGENT IDENTIFICATION IS REQUESTED, PLEASE PROVIDE A BRIEF EXPLANATION UNDER "REMARKS".

10. HOST INFORMATION  
 NAME OF HOST (Scientific name when possible)  
 Solanum tuberosum

11. QUANTITY OF HOST PLANTS AFFECTED (Insert figure and indicate  Number  Percent):  
 15 acres

12. PLANT DISTRIBUTION  
 LIMITED  
 SCATTERED  
 WIDESPREAD

13. PLANT PARTS AFFECTED  
 Leaves, Upper Surface  Trunk/Bark  Bulbs, Tubers, Corms  Seeds  
 Leaves, Lower Surface  Branches  Buds  
 Petiole  Growing Tips  Flowers  
 Stem  Roots  Fruits or Nuts

14. PEST DISTRIBUTION  
 FEW  
 COMMON  
 ABUNDANT  
 EXTREME

15.  INSECTS  NEMATODES  MOLLUSKS

NUMBER SUBMITTED	LARVAE	PUPAE	ADULTS	CAST SKINS	EGGS	NYPHPS	JUVS.	CYSTS
ALIVE								
DEAD								

16. SAMPLING METHOD  
 Soil sample

17. TYPE OF TRAP AND LURE  
 Samples

18. TRAP NUMBER  
 Soil Samples 16, 18, 19, 20, 30

19. PLANT PATHOLOGY - PLANT SYMPTOMS ("X" one and describe symptoms)  
 ISOLATED  GENERAL

20. WEED DENSITY  
 FEW  SPOTTY  GENERAL

21. WEED GROWTH STAGE  
 SEEDLING  VEGETATIVE  FLOWERING/FRUITING  MATURE

22. REMARKS  
 PROMPT determination requested; detection outside quarantine area.  
 New Township Record, Fremont Township, Steuben County, New York

23. TENTATIVE DETERMINATION  
 50 Globodera rostochiensis cysts, viable and nonviable. DET: E. N. Jones

24. DETERMINATION AND NOTES (Not for Field Use)

**FOR IIBIII USE**  
 DATE RECEIVED  
 NO. LABEL SORTED PREPARED  
 DATE ACCEPTED  
 RR

SIGNATURE \_\_\_\_\_ DATE \_\_\_\_\_

**PPQ FORM 391 (AUG 02)** Previous editions are obsolete.

This is a 6-Part form. Copies must be disseminated as follows:  
 PART 1 - PPQ  PART 2 - RETURN TO SUBMITTER AFTER IDENTIFICATION  PART 3 - IIBIII OR FINAL IDENTIFIER  
 PART 4 - INTERMEDIATE IDENTIFIER  PART 5 - INTERMEDIATE IDENTIFIER  PART 6 - RETAINED BY SUBMITTER

FIGURE A-1-12 Example of PPQ Form 391, Specimens for Determination

## Purpose

PPQ Form 391, *Specimens for Determination*, is used to submit along with collections from golden nematode survey for identification. In addition to the Golden Nematode Program, PPQ Form 391 is also used for other domestic collections (other special survey programs, export certification, local and individual collections, and warehouse inspections).

## Instructions

Complete PPQ Form 391 following the instructions in [Table A-1-9](#).

**TABLE A-1-9 Instructions for Completing PPQ Form 391, Specimens for Determination**

Block		Instructions
1	COLLECTION NUMBER	<p>1. ASSIGN a number for each collection as follows:            Last 2 digits of current year-collector's initials'-3-digit number, starting with -001 and continue consecutive numbering for each subsequent collections</p> <div style="border: 1px solid black; padding: 5px; margin: 5px 0;"> <p><b>EXAMPLE</b> In 2005, Samuel L. Jones collected his first specimen for determination of the year. His first collection number is 05-SLJ-001.</p> </div> <p>2. ENTER the collection number</p>
2	DATE	ENTER the date
3	SUBMITTING AGENCY	PLACE an X in the appropriate block (State Cooperator/PPQ/Other); if other, list the Agency
4	NAME OF SENDER	LIST your (sender/collector's) name
5	TYPE OF PROPERTY	LIST the type of property (where specimen collected-farm, feedmill, nursery, etc.)
6	ADDRESS OF SENDER	List your (sender/collector's) address
7	NAME AND ADDRESS OF PROPERTY OR OWNER	List the name and address of the property (where specimen collected)
8A-8H	REASONS FOR IDENTIFICATION	PLACE an X in the appropriate blocks
9	IF PROMPT OR URGENT IDENTIFICATION IS REQUESTED, PLEASE PROVIDE A BRIEF EXPLANATION UNDER "REMARKS"	Leave blank; enter any remarks in <i>Block 22</i>
10	HOST INFORMATION	If possible, ENTER the scientific name of the host
11	QUANTITY OF HOST	If applicable, ENTER the quantity of host and plants affected
12	PLANT DISTRIBUTION	If applicable, check appropriate boxes
13	PLANT PARTS AFFECTED	If applicable, check appropriate boxes
14	PEST DISTRIBUTION	PLACE an X in the appropriate box (FEW, COMMON, ABUNDANT, EXTREME)

**TABLE A-1-9 Instructions for Completing PPQ Form 391, Specimens for Determination (continued)**

Block		Instructions
15	INSECTS/NEMATODES/ MOLLUSKS	1. PLACE an X in the box to indicate type of specimen 2. ENTER the number of specimens submitted
16	SAMPLING METHOD	LIST the sampling method
17	TYPE OF TRAP AND LURE	LIST the type of trap and lure used
18	TRAP NUMBER	LIST the soil sample numbers
19	PLANT PATHOLOGY-PLANT SYMPTOMS	If applicable, check appropriate box
20	WEED DENSITY	Leave blank
21	WEED GROWTH STAGE	Leave blank
22	REMARKS	If PROMPT or URGENT identification is required, indicate and give a brief explanation
23	TENTATIVE DETERMINATION	LIST any tentative determination made
24	DETERMINATION AND NOTES (Not for Field Use)	Leave blank (completed by final identifier)

### Distribution

Distribute PPQ Form 391 as follows:

1. Send a copy to the golden nematode identifier.
2. Place a copy in the program file.
3. If confirmed as a non-infested field (negative survey), file in the county folder.
4. If the field is numbered with a Roman numeral, file in that folder (Upstate NY).
5. If confirmed as an infested field (positive survey), then attach to PPQ Form 312 and file in the infested field folder.



4.) All soil residues shall be treated with on of the schedules below:

a. DRY HEAT:

<u>Temperature</u> Celcius / Farenheit	<u>Exposure Period</u>
110 - 120.5 C (230-249 F)	16 hours
121 - 154 C (250-309 F)	2 hours
154.5 - 192.5 C (310-379 F)	30 minutes
193 - 220 C (380-429 F)	4 minutes
221 - 232 C (430-450 F)	2 minutes

Do not start counting time until the entire mass reaches the required temperature, or:

b. Steam Heat:

15 pounds pressure for 30 minutes.

Individual packages of 5 pounds or less or, if in trays, the soil residues should not exceed 2 inches in depth.

Do not start counting time until pressure reaches 15 pounds.

c. Any other procedure must first be approved by the Deputy Administrator, USDA, APHIS PPQ

5. Any water residues (effluent) from the processing of soil samples must be treated by one of the following approved schedules before discarding so as not to present a hazard of pest spread:

a. Domestic and Foreign - Boil the effluent for 1 minute, or;

b. Domestic only - Filter through a 100 mesh screen. The residues left in the filter should be burned.

Initial XW

FIGURE A-1-14 Example of Attachment to Compliance Agreement for Potato Growers (page 2 of 4)

### **GOLDEN NEMATODE RESEARCH FARM SAFEGUARDS**

**APHIS and ARS personnel have developed the following safeguards to be enforced at the Nematode Research Farm, Steuben County, New York.**

- 1.) The field will be enclosed within an approved fence. A steel gate at the entrance to the fenced area shall be kept locked when farm is unattended. At the beginning of the approach to the farm, a chain barrier shall be maintained across the road and this, likewise, shall be kept locked.
- 2.) Only authorized personnel are allowed entry to the fenced area.
- 3.) The fenced areas will be posted with proper identification to discourage unauthorized entry.
- 4.) A supply of plastic boots shall be maintained inside the entrance of the fenced area for use by authorized visitors. A small building shall be erected just inside the entrance to the fenced area to store plastic boots and other sanitation equipment.
- 5.) Vehicles used by personnel going to the research farm shall be parked outside the fenced area.
- 6.) Vehicles and other equipment used within the fenced area shall be stored within the fenced area. Any equipment, vehicles, or supplies removed from the fenced area shall be decontaminated by removal of excess soil through the use of steam or water and fumigated at approved schedules in the paved decontamination area designated for this purpose.
- 7.) A minimum 50-foot-wide border of sod shall encompass the cultivated portion of the research farm within the fenced area. If necessary, based on land topography and utilization, an additional sod area will be maintained outside the fenced area.
- 8.) A runoff control structure shall be maintained to contain all runoff from the field-plot research area. No field-plot research will be done outside breakpoint to the runoff control structure.
- 9.) All water runoff from the main building area (laboratory, shop, and equipment storage) shall be contained within the fenced area by a series of diversion ditches and a drywell. Runoff from the road (from decontamination area to building) shall be likewise contained.

Initial XW

**FIGURE A-1-15 Example of Attachment to Compliance Agreement (page 3 of 4)**

## SAFEGUARDS

- 10.) Water from the decontamination area shall be disposed of through a system consisting of a drywell inside the fence and a drain field just outside the fence. This system will be treated periodically with a nematicidal chemical as the need warrants.
- 11.) To reduce contamination, the entire area around the main building consisting of 21,000 sq. ft. shall be covered with 4-6 inches of crushed stone. In addition, the road from the decontamination area to the building (7,000 sq. ft.) shall be likewise covered with crushed stone. This crushed stone area shall be treated periodically with a nematicidal chemical as the need warrants.
- 12.) Drivers and vehicles for necessary deliveries shall be confined to the crushed stone area. This area shall be treated with a nematicidal chemical just prior to deliveries. Delivery vehicles confined to the crushed stone area shall be decontaminated by steam. Decontamination of other delivery vehicles shall be approved by APHIS-PPQ personnel, depending upon nature of delivery. Delivery shall be kept to a minimum. Potato seed storage shall be constructed to eliminate potato seed delivery to the farm.
- 13.) Personnel working within the fenced area shall wear outer clothing and shoes which shall be left within the fenced area. Such clothing shall be steam sterilized before released from the research project.
- 14.) Soil samples or other material transported from the research farm shall be carried in leak proof containers under safeguards to prevent spillage.
- 15.) All potatoes grown on the research farm, except those required for experimental purposes, shall be disposed of inside the fenced area.
- 16.) Operations will be subject to periodic inspection by APHIS-PPQ or New York Department of Agriculture and Markets personnel for adherence to safeguards, and adjustments made as necessary.

Initial XW

FIGURE A-1-16 Example of Attachment to Compliance Agreement (page 4 of 4)

## Purpose

PPQ Form 519, Compliance Agreement, is completed by the PPQ Office/ Plant Health Safeguarding Specialist for the area of coverage.

## Instructions

Complete PPQ Form 519 as shown in [Table A-1-10](#).

**TABLE A-1-10 Instructions for Completing PPQ Form 519, Compliance Agreement**

Block		
1	NAME AND ADDRESS OF PERSON OR FIRM	ENTER the name and address of the person or firm with whom the agreement is made
2	LOCATION	ENTER the location of the land
3	REGULATED ARTICLES	ENTER the name of the regulated article  <b>EXAMPLE</b> <span style="border: 1px solid black; padding: 2px;">Soil samples for analysis.</span>
4	APPLICABLE FEDERAL QUARANTINE(S) OR REGULATIONS	ENTER GOLDEN NEMATODE 7CFR§301.85
5	I/we agree to the following	LIST the terms of the agreement
6	SIGNATURE	Person authorizing agreement signs
7	TITLE	ENTER your title
8	DATE SIGNED	ENTER the date signed
9	AGREEMENT NO.	ENTER the agreement number
10	DATE OF AGREEMENT	ENTER the date the agreement is made
11	PPQ OFFICIAL	ENTER the name and address of the PPQ Official authorized to execute the agreement
12	ADDRESS	ENTER the address and telephone number  U.S. Department of Agriculture, APHIS Plant Protection and Quarantine 8237 Kanona Road Avoca, NY 14809 607/566-2212
13	SIGNATURE	PPQ Official SIGNS and lists title
14	STATE AGENCY OFFICIAL	ENTER the name and address of the State agency official entering the agreement

**TABLE A-1-10 Instructions for Completing PPQ Form 519, Compliance Agreement**

Block		
15	ADDRESS	ENTER the address of the State Agency New York State Department of Agriculture and Markets Division of Plant Industry PO Box 57 Little Valley, NY 14755
16	SIGNATURE	State agency official SIGNS

### **Distribution**

Distribute PPQ Form 519 and attachments, as follows:

- ◆ Copy to person signing the agreement
- ◆ Copy to the State Official
- ◆ Place the original in the PPQ County file Compliance Agreements folder

**PPQ Form 530, Limited Permit**

No. 444576

**U.S. DEPARTMENT OF AGRICULTURE  
 ANIMAL AND PLANT HEALTH INSPECTION SERVICE  
 PLANT PROTECTION AND QUARANTINE PROGRAMS  
 HYATTSVILLE, MARYLAND 20782  
**LIMITED PERMIT****

*This permit must be surrendered to the consignee at destination of shipment.*

This permit authorizes the movement of the **NONCERTIFIED** articles described below to a specified destination for limited handling, utilization, or processing, or for treatment. The movement of such articles is regulated by Federal or State cooperative domestic plant quarantines.

1. DATE ISSUED	2. VOID AFTER
3. NAME OF CONSIGNOR	
4. SHIPPING POINT	
5. NAME AND ADDRESS OF CONSIGNEE	
6. VEHICLE LICENSE NO. & STATE	7. R. R. CAR INITIALS & NO.

8. DESCRIPTION		
QUANTITY A	ARTICLE B	REMARKS C

9. SIGNATURE OF ISSUING INSPECTOR

**ENDORSEMENT**

*The above described shipment was received by the designated consignee, and was handled in the manner approved under the provisions of all applicable Federal or State cooperative domestic plant quarantines.*

10. DATE RECEIVED

11. SIGNATURE OF DESTINATION INSPECTOR

**PENALTY FOR MISUSE OR ALTERATION**

(7-USC-163)

PPQ FORM 530 (10/73)  
 REPLACES PPD FORM 5-3 (9/70), WHICH  
 MAY BE USED

CONSIGNEE'S COPY

**FIGURE A-1-17 Example of PPQ Form 530, Limited Permit (blank)**

## Purpose

PPQ Form 530, Limited Permit, is used to authorize movement of **noncertified** regulated articles to a specific approved destination for processing and treatment.

## Instructions



**Important**

Movement of noncertified regulated articles **must** be approved by the appropriate State Regulatory Official in the applicable State and/or the PPQ Deputy Administrator or designee.

See **Special Procedures** on page 3-1-4. Complete PPQ Form 530 as shown in **Table** below.

**TABLE A-1-11 Instructions for Completing PPQ Form 530, Limited Permit**

Block		Instructions
1	DATE ISSUED	
2	VOID AFTER	
3	NAME OF CONSIGNOR	
4	SHIPPING POINT	
5	NAME AND ADDRESS OF CONSIGNEE	
6	VEHICLE LICENSE NO. & STATE	
7	R.R. CAR INITIALS & NO.	
8	DESCRIPTION	
	QUANTITY A	
	QUANTITY B	
	QUANTITY C	
9	SIGNATURE OF ISSUING INSPECTOR	
10	DATE RECEIVED	
11	SIGNATURE OF DESIGNATION INSPECTOR	

## Distribution

Distribute PPQ Form 530, Limited Permit, as follows:

- ◆ XX
- ◆ XX

## PPQ Form 540, Certificate of Federal/State Domestic Quarantines

992002

U.S. DEPARTMENT OF AGRICULTURE  
 Animal and Plant Health Inspection Service  
 Plant Protection and Quarantine Programs  
 Hyattsville, Maryland 20782

**CERTIFICATE**

*This certificate must be surrendered to the  
 consignee at destination of shipment.*

The articles described below are certified under all applicable  
 Federal or State cooperative domestic plant quarantines.

1. DATE ISSUED	2. VOID AFTER
----------------	---------------

3. NAME OF CONSIGNOR \_\_\_\_\_

4. SHIPPING POINT \_\_\_\_\_

5. NAME & ADDRESS OF CONSIGNEE \_\_\_\_\_  
 \_\_\_\_\_  
 \_\_\_\_\_

6. VEHICLE LICENSE NO. & STATE	7. R. R. CAR INITIALS & NO.
--------------------------------	-----------------------------

8. DESCRIPTION		
QUANTITY A	ARTICLE B	REMARKS C

9. SIGNATURE OF ISSUING INSPECTOR \_\_\_\_\_

**PENALTY FOR MISUSE OR ALTERATION  
 (7-USC-163)**

PPQ FORM 540 (11/76)  
 PPQ FORM 540 (1/73) MAY BE USED

ISSUING INSPECTOR'S COPY

**FIGURE A-1-18 Example of PPQ Form 540, Certificate of Federal/State Domestic Quarantines**

**Purpose**

PPQ Form 540, Certificate of Federal/State Domestic Quarantines, is used by the Golden Nematode Program to certify that the regulated articles (listed on the form) are certified free from golden nematode only.

**Instructions**

Complete PPQ Form 540 as shown in [Table A-1-12](#).

**TABLE A-1-12 Instructions for Completing PPQ Form 540, Certificate of Federal/State Domestic Quarantines**

Block		Instructions
1	DATE ISSUED	
2	VOID AFTER	
3	NAME OF CONSIGNOR	
4	SHIPPING POINT	
5	NAME AND ADDRESS OF CONSIGNEE	
6	VEHICLE LICENSE NO. & STATE	
7	R.R. CAR INITIALS & NO.	
8	DESCRIPTION	
	QUANTITY A	
	QUANTITY B	
	QUANTITY C	

**Distribution**

Distribute PPQ Form 540, Certificate of Federal/State Domestic Quarantines, as follows:



# B

Golden Nematode  
Program Manual

## Appendix B

### *Emergency Aid and Safety*

---

#### Contents

Introduction	page B-1-1
Fumigant Safety Precautions	page B-1-1
Fumigant Monitoring	page B-1-3
Emergency Action	page B-1-4
First Aid Rescue Breathing	page B-1-5
Signs and Symptoms of Poisoning and Emergency Aid and Medical Treatment for Some Fumigants Used by APHIS	page B-1-6

---

#### Introduction

The *Emergency Aid and Safety* appendix covers emergency precautions, aid, and safety for some fumigants. This appendix is reprinted from the USDA-APHIS-PPQ *Treatment Manual*.

---

#### Fumigant Safety Precautions

Fumigants are generally toxic when inhaled or spilled on skin or clothing.

You carefully **must** read the label of each product used in the treatment of golden nematode before using the product. Review ***Fumigant Monitoring*** before applying any pesticide. This information is also available in the *PPQ Treatment Manual*.

If an accident should occur immediately follow and implement the First Aid measures listed on the label of the pesticide being used.

#### **Safety Procedures**

Follow specific precautions listed on the label and labeling of each fumigant to be used.

Hazards of each fumigant vary with the following:

- ❖ Relative toxicity of each fumigant
- ❖ Dosage rate and concentration
- ❖ Enclosure size
- ❖ Enclosure tightness
- ❖ Physical condition of employee (allergies, heart condition, respiratory ailments, etc.)

### **Guidelines for Using Fumigants Safety**

Review and follow the guidelines below:

1. Know the characteristics of the fumigants you are working with.
2. Have the proper equipment to carry out the fumigation.
3. Be familiar with the emergency aid that would be required should an accident occur.
4. If there is any chance of exposure to highly-toxic fumigants, then wear protective equipment as follows:
  - ❖ Face shield or respirator should be used when liquids are being transferred and there is a possibility of splattering
  - ❖ Gloves should be impermeable to the liquid fumigant being used
  - ❖ Rubber aprons should be long enough to prevent legs from being exposed
5. Dispensers for measuring the amount of fumigant should have shatter-proof shields.
6. The area surrounding the fumigation enclosure should be well-aerated. Operators should be located upwind from treatment.
7. If necessary to stay in the treatment area, the air should be monitored to determine if harmful levels of fumigant are present.
8. Under **no** circumstance should an inspector be exposed to concentrations above minimum safe standards.
9. A self-contained breathing apparatus (SCBA) is required at all fumigation sites and should always be readily available in case an emergency develops.
  - A. You must have a medical evaluation and clearance to use SCBA equipment. The evaluation must be performed by a physician or licensed health care professional.
  - B. You must follow OSHA standards for respirator use (see *APHIS Health and Safety Manual*, Chapter 11, Section 3).

10. Use of SCBA respirators are mandatory for all PPQ Officers within 30 feet of tarpaulin fumigation or when TLV is exceeded (5 ppm for methyl bromide).
11. Persons working regularly with toxic fumigants should have blood tests and physical examinations if warranted by supervisor's consultation with local medical authorities.
12. A first-aid kit equipped with the proper materials should be readily available at the treatment site.
13. Telephone numbers of local hospitals, doctors, and poison control centers should be prominently displayed.
14. Learn to recognize the signs and symptoms of fumigant poisoning. Training should be given to each inspector.
15. Supervisors should be aware of the signs of fatigue. The risk of accidents increases in tired employees.
16. **Do not** eat, drink, smoke, or carry tobacco in areas where fumigants are being used.

## Fumigant Monitoring

Fumigant	Route of Entry	Detector Unit or Monitoring Device	Source of Exposure
Chloropicrin	Inhalation	None	Application of liquid, leakage from enclosure, aeration
Methyl bromide	Inhalation; skin	Gas detector tubes, Halide detector, T/C unit	Cylinder connection, leaks in tarpaulin, applicators, aeration
Phosphine (from aluminum phosphide)	Inhalation	Gas detector tubes	Application of pellets, leakage from enclosure, aeration
Sulfuryl fluoride (Vikane)	Inhalation	T/C Unit	Applicator and cylinder connections, leakage from enclosure, aeration

**FIGURE B-1-1 Fumigant Route of Entry, Monitoring Device, and Exposure Source**

## Emergency Action

### Self (You)

If you are exposed to a fumigant, immediately move away from the contaminated area. Notify your co-workers of the danger and that you have been exposed. Onset symptoms may be delayed with some fumigant. Notify your supervisor promptly of the details.

If liquid fumigants have spilled on your skin or clothing, immediately remove the contaminated clothing and gently wash your skin with large quantities of soap and water. **Do not** use abrasive cloths or brushes. Be sure to clean the area under your fingernails and toenails with soap and water. You may also rinse contaminated skin with rubbing alcohol.



Dangerous vapors will be produced by the liquid fumigant during evaporation from skin and clothing.

### Contaminated Clothing

After you have removed your contaminated clothing, be sure **not** to use or wear the clothing again until thoroughly aired, washed, dried. Properly dispose of any clothing that has been damaged by the fumigant.

### Co-worker

If chemical intoxication due to exposure is suspected at any time do as follows:

1. Immediately move the victim out of the exposed area and into fresh air.



**Do not** enter a contaminated area without a proper respirator, even to effect rescue.

2. If there is evidence of respiratory weakness, then give artificial respiration. Oxygen can be beneficial. Artificial respiration, when needed, takes precedence over all other first aid (see **First Aid Rescue Breathing**).
3. If symptoms suggest immediate care is needed, then call a physician.
4. Keep the patient warm, comfortable, and quiet as possible.
5. If convulsions occur, use gentle restraint to prevent injury.

## First Aid Rescue Breathing

If you believe a person has stopped breathing, give First Aid rescue breathing immediately. Ask someone else to get medical help.

### 1. Is the person breathing?



To find out, place the person on his/her back and put your ear close to his/her mouth. If the person is breathing you will hear his breath and see his chest rise and fall. If the person is not breathing, continue to 2., open the airway.

### 2. Open the airway.



If the person has stopped breathing, lift up his/her neck with one hand and push down on the person's forehead with the other hand. This opens the airway and the person may start to breathe. If the person doesn't breathe, then begin rescue breathing at once.

### 3. Start rescue breathing.



Keep one hand under the person's neck so that his/her chin is tilted backward and the chin is up. Pinch the nostrils shut using the fingers of your other hand. Take a deep breath and cover the person's mouth completely with your own. Blow air into his/her mouth. When the person's chest moves up, move your mouth away and let the person's chest go down by itself. Repeat this procedure every 5 seconds. Do not stop until the person starts breathing or medical help arrives.

**FIGURE B-1-2 First Aid Rescue Breathing**

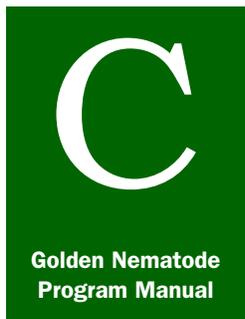
**Appendix B:**

Signs and Symptoms of Poisoning and Emergency Aid and Medical Treatment for Some Fumigants Used by APHIS

**Signs and Symptoms of Poisoning and Emergency Aid and Medical Treatment for Some Fumigants Used by APHIS**

Fumigant	Signs and Symptoms	Emergency Aid	Medical Treatment
Chloropicrin	<ul style="list-style-type: none"> <li>◆ Powerful irritant that affects all body surfaces</li> <li>◆ Lacrimation, vomiting, bronchitis, pulmonary edema</li> <li>◆ Inhalation causes anemia, weak and irregular heartbeat, recurrent asthmatic attacks</li> </ul>	<ul style="list-style-type: none"> <li>◆ Artificial respiration</li> <li>◆ Oxygen if available</li> </ul>	<p>Symptomatic—oxygen</p> <p>Sample analysis might be helpful in diagnosis and prognosis</p>
Methyl Bromide	<ul style="list-style-type: none"> <li>◆ Central nervous system depression</li> <li>◆ Nausea, fever, pulmonary edema</li> <li>◆ Confusion, delirium, mania, staggering, tremors, visual disturbances</li> <li>◆ Abdominal pain, convulsions, coma</li> <li>◆ <b>Onset may be delayed 4-12 hours</b></li> <li>◆ On skin, severe irritation, blisters, dermatitis</li> </ul>	<ul style="list-style-type: none"> <li>◆ Artificial respiration</li> <li>◆ Oxygen if available</li> <li>◆ <b>Do not</b> use mechanical resuscitation</li> <li>◆ If fumigant contacts skin, wash 15 minutes with large amounts of water</li> <li>◆ If fumigant contacts clothing, vapors may be released in toxic quantities</li> </ul>	<p>Symptomatic—artificial respiration</p> <p>Analysis of breath and blood may help in diagnosis and prognosis</p> <p>For nausea accompanied by vomiting, give intravenous glucose-bearing vehicles</p>
Phosphine (from aluminum phosphide)	<ul style="list-style-type: none"> <li>◆ 2,000 ppm in air is rapidly fatal; death may be delayed several day</li> <li>◆ Chest pain, headache, dyspnea, restlessness, vomiting</li> <li>◆ Convulsions, coma, paralysis</li> <li>◆ Low blood pressure, slow heart</li> </ul>	<ul style="list-style-type: none"> <li>◆ Artificial respiration</li> <li>◆ Oxygen if available</li> </ul>	<p>Symptomatic—oxygen</p> <p>Control convulsions with sedatives</p> <p>Restore fluid balance with glucose and saline</p>
Sulfuryl Fluoride	<p>Central nervous system depression; excitation may follow</p>	<p>Place patient in fresh air, face downward, with head slightly below level of lungs, and keep warm</p> <p>If breathing stops, give artificial respiration</p>	<p>First symptoms expected are those of respiratory irritation and central nervous system depression—treat symptomatically</p>

**FIGURE B-1-3 Signs and Symptoms of Fumigant Poisoning and Emergency Aid**



# Appendix C

## *Maintenance of the Manual*

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### Contents

Introduction	page C-1-1
Policy for Maintaining the Golden Nematode Manual	page C-1-1
Revisions to Manuals	page C-1-2
Advance Notices	page C-1-2
Immediate Updates	page C-1-2
New Editions	page C-1-2
Supersede Statements	page C-1-3
Keeping the Manual Current	page C-1-3
Control Data	page C-1-3
Transmittal Memos	page C-1-3
Update Record	page C-1-3
Responsibilities of Manual Users	page C-1-4
Address and Copy Count Changes	page C-1-4
Additional Manuals and Revision Copies	page C-1-5

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### Introduction

This appendix contains information about revisions to manuals and the procedures and responsibilities for users to maintain the *Golden Nematode Manual*. Most manuals are available electronically at the PPQ Manuals Unit Web site at the following URL:

<[http://www.aphis.usda.gov/ppq/manuals/online\\_manuals.html](http://www.aphis.usda.gov/ppq/manuals/online_manuals.html)>

Some manuals are also issued in paper (hard) copy.

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### Policy for Maintaining the *Golden Nematode Manual*

This appendix contains the systematic procedures and the users' responsibilities for maintaining this manual. How many manuals are supplied to each separate work location varies according to the needs of the location or designated through channels. Each work location **must** have at least one designated individual to maintain each manual. Failure to maintain each manual according to procedures prescribed herein is likely to substantially reduce the effectiveness of the programs the manuals support.

## Revisions to Manuals

PPQ's Manuals Unit issues revisions by e-mail transmittal memo that identify and either contain an Adobe Acrobat pdf copy of the pages to be added or replaced or refer the manual user to the Manuals Unit Web site to download a pdf copy the updated section of the manual. In addition to e-mail, paper (hard copy) transmittal memos and revisions are occasionally issued. New pages are **not** issued solely to correct a minor typographical error; however, errors that would lead to an incorrect action are immediately corrected and updated.

### Advance Notices

Advance notices are issued when the revisions are **not** urgent or are extensive (more than 6 pages).

Transmittals (by e-mail or memorandum) for advance notices usually contain the following information:

- ◆ Transmittal number (used to track updates)
- ◆ Alert to users that the manual has been updated
- ◆ Purpose of the revision
- ◆ Supersede statement
- ◆ Instructions to add new pages and remove old pages and replace with new ones

### Immediate Updates

Immediate updates are issued when the revisions are urgently needed and cover 6 pages or less.

Transmittals (by e-mail) for immediate updates usually contain the following information:

- ◆ Transmittal number (used to track updates)
- ◆ Purpose of the immediate update
- ◆ Supersede statement
- ◆ List of the updated pages
- ◆ Instructions to add new papers and to remove old pages and replace with new ones
- ◆ Attachment containing the updated pages

### New Editions

New editions of manuals are scheduled at fixed intervals—barring budgetary constraints—at least every 4 years or when the percentage of updated pages exceeds 30 percent of the entire manual.

### Supersede Statements

Supersede statements identify existing official documents that are no longer valid or accurate because they have been incorporated into a manual. Supersede statements are included in transmittals (e-mail or memorandum) and are critical from a legal standpoint.

PPQ's Manuals Unit is obligated to identify all existing official documents that are superseded. If manual users accessed outdated information that was **not** formally superseded, then the Agency may be liable.

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### Keeping the Manual Current

There are three ways to track revisions to your manual: control data, transmittal memos, and the **Update Record**.

#### Control Data

The manual has control data positioned at the bottom of most pages. The control data contains the month, year, and transmittal number. New editions of manuals **always** start with -01. The transmittal number increases by 1 for each manual update.

<b>EXAMPLE</b>	07/2005-01 PPQ ◆ 07/2005 is the month and year the manual was issued ◆ -01 is the transmittal number
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#### Transmittal Memos

All new editions and updates are accompanied by transmittal memos that list the transmittal number. Work locations are required to numerically file all transmittal memos. The filed transmittals will be used by Headquarters to audit manuals during port reviews. You may also want to individually retain and file transmittals to assure yourself that you have received all previous issuances.

#### Update Record

The **Update Record** is located the page following the title page of the manual. For new editions issued on hard copy (paper), the **Update Record** is on the reverse side of the title page. If you **do not** already have a hard copy of this page, you can get a printed copy from the Web site.

Record the transmittal number and the date each transmittal memo update is received on the **Update Record**. This will let you help you identify if you are missing any transmittals—especially if you find the transmittal numbers are out-of-sequence. During port reviews and/or audits, this record shows if the work location manual is up-to-date.

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## Responsibilities of Manual Users

Electronic copies of each manual are revised and maintained by the Manuals Unit and are available on PPQ's Manuals Unit Web site.

If your work location requires that you print and maintain a hard copy from the posted electronic copy, then continue below.

### Maintain the Paper Copy

To maintain the *Golden Nematode Manual* paper (hard) copy effectively and to enhance professionalism, the manual **must** be kept up-to-date. If you fail to keep the manuals at your work location updated, you run the risk of making a decision that provides the opportunity for an outbreak of an exotic disease. Your diligence is a crucial part of this process.

When you receive each transmittal and update, do the following:

1. Read the transmittal (memorandum or e-mail) and pages that are part of the transmittal to understand the purpose of the revision. Except for changes to the Index, List of Figures, and List of Changes, all revisions are marked with a change bar as located to the left of this sentence.
2. Record the transmittal number and date the transmittal and was received in the **Update Record**.
3. Add new pages or remove and replace with the revised pages on the same day you receive the transmittal.
4. If required locally, numerically file the transmittal e-mail or memorandum.
5. If you have missed a transmittal, check the Manuals Unit Web site first.

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## Address and Copy Count Changes

Paper copies of some manuals are mailed from the APHIS Printing, Distribution, and Mail Branch located in Riverdale, Maryland. If you receive a paper (hard) copy, then be sure to communicate, through proper channels, changes to address and copy counts for the mailing list.

When updating mailing lists, always provide the following information:

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FAX: 301-734-8455  
E-mail: <edward.s.lawson@aphis.usda.gov>

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### **Additional Manuals and Revision Copies**

If the manual and revisions are issued in paper (hard) copy, then you may order additional manuals or revisions. Always provide the following information:

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- ◆ Contact person's name, phone number, and FAX number
- ◆ Title of manual
- ◆ Transmittal number (if known)
- ◆ Number of copies needed

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FAX: 301-734-8455  
E-mail: <mary.l.kellington@aphis.usda.gov>



# Glossary

## Definitions, Terms, and Abbreviations

**certificate.** A document issued or authorized to be issued...by an inspector to allow the interstate movement of regulated articles to any destination.

**compliance agreement.** A written agreement between a person engaged in growing, handling, or moving regulated articles, and the Plant Protection Programs, wherein the former agrees to comply with the requirements of this subpart identified in the agreement by the inspector who executes the agreement on behalf of the Plant Protection and Quarantine Programs as applicable to the operations of such person.

**exposure period.** Time required for the soil to become free of fumigant. The duration of the exposure period is influenced by both temperature and soil moisture. The higher the temperature, the shorter the fumigation time. The cooler the temperature, the longer the fumigation time.

**delimiting survey.** Survey conducted to establish the boundaries of golden nematode infestation in a field or property.

**detection survey.** Survey conducted to determine whether a field or land is infested with golden nematode.

**farm tools.** An instrument worked or used by hand; e.g., hoes, rakes, shovels, axes, hammers, and saws.

**fluming.** An old cleaning process where potatoes are run down a flume to remove soil from the potatoes without the use of water. **Not** commonly used today, but still an option for growers to use.

**generally infested area.** Any part of a regulated area **not** designated as a suppressive area in accordance with 301.85.2

**inspector.** Any employee of the Plant Protection and Quarantine Programs, Animal and Plant Health Inspection Service, U.S. Department of Agriculture, or other person, authorized by the Deputy Administrator to enforce the provisions of the Quarantine and regulations...

**interstate.** From any State into or through any other State.

**limited permit.** A document issued or authorized to be issued by an inspector to allow the interstate movement of noncertifiable regulated articles to a specified destination for limited handling, utilization or processing for treatment.

**mechanized cultivating equipment.** Mechanized equipment used for soil tillage, including tillage attachments for farm tractors (tractors, disks, plows, harrows, planters, and subsoilers).

**mechanized harvesting equipment.** Mechanized equipment used for harvesting purposes (combines, potato conveyors, harvesters, and hay balers).

**Glossary:**Definitions, Terms, and Abbreviations

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**mechanized soil moving equipment.** Equipment used for moving or transporting soil (draglines, bulldozers, dump trucks, road scrapers, etc.).

**mineral soil.** Soil consisting primarily of mineral (sand, silt, and clay) material.

**monoculture.** A single crop planted on a farm or in a region or county.

**moved, movement, move.** Shipped, deposited for transmission in the mail, otherwise offered for shipment, received for transportation, carried, or otherwise transported, or moved, or allowed to be moved, by mail or otherwise.

**muck soil.** Soil consisting primarily of organic matter.

**person.** Any individual, corporation, company, society, or association, or other organized group of any of the foregoing.

**regulated area.** Any quarantined State, or any portion thereof, listed as a regulated area in 7CFR §301.85-2(a) or otherwise designated as a regulated area in accordance with §301.85-2(b).

**regulated article.** Any articles as described in xx.

**resistant variety treatment.** Planting a golden nematode resistant variety of potatoes which controls the golden nematode population in the same manner as a chemical treatment. Although the nematode still is caused to hatch by the resistant potato plants, the nematode is unable to survive.

**restricted destination permit.** a document issued or authorized to be issued by an inspector to allow the interstate movement of regulated articles not certifiable under all applicable Federal domestic plant quarantines to a specified destination for other than scientific purposes.

**Ro1.** Traditional golden nematode strain.

**Ro2.** New golden nematode biotype.

**sample.** In the Golden Nematode Program, a small portion of soil (mineral or muck in New York) that is collected for processing.

**scientific permit.** A document issued by the Deputy Administrator to allow the interstate movement to a specified destination of regulated articles for scientific purposes.

**soil.** That part of the upper layer of earth in which plants can grow.

**State.** Any State, territory, or district of the United States, including Puerto Rico.

**suppressive area.** That portion of a regulated area where eradication of infestation is undertaken as an objective as designated under §301.85-2(a).

**survey.** xx.

**T/C Unit (thermal conductivity).** device used to measure gas concentration levels in tarpaulins and chambers.

**Tier.** length of an edge of the field in a straight line.

# Index

## A

Agricultural land  
Category 3 [3-1-27](#)

Aluminum phosphide [B-1-3](#)

Articles [3-1-2](#)

## B

Bags  
labeling example [2-2-17](#)  
labeling instructions [2-2-16](#)  
new [3-1-16](#)

Beets, sugar [3-1-22](#)

Bench soil [3-1-24](#)

Biometrics survey [2-3-14](#)

Block method [2-3-8](#)

Boxcars [3-1-16](#)

Boxes, used [3-1-3](#)

Bulbs [3-1-3](#), [3-1-21](#)

Bulk soil [3-1-23](#)

Burlap  
bags [3-1-3](#)  
containers [3-1-16](#)

## C

Canada [3-1-4](#)

Carriers [2-2-18](#), [2-3-4](#)

Category 1 Non-agricultural land [3-1-27](#)

Category 2 Non-agricultural land [3-1-27](#)

Category 3 agricultural land [3-1-27](#)

Certificates  
determine if required [3-1-7](#)  
regulated articles [3-1-2](#)

Certification period [3-1-19](#), [3-1-26](#)

Chemicals  
exudates [2-1-5](#)

Chloropicrin [B-1-3](#)

Cleaning  
clothing and equipment [2-2-18](#), [2-3-5](#)  
confirmatory surveys [2-2-20](#), [2-3-5](#)  
other surveys [2-2-20](#), [2-3-5](#)

Cloth containers [3-1-16](#)

Commercial potato fields [2-2-18](#), [2-3-5](#)

Compliance agreements [3-1-26](#)

Compost  
regulated article [3-1-2](#)

Confirmation survey [2-3-12](#)

Consumer packages [3-1-16](#)

Containers  
approved [3-1-16](#)  
movement of used [3-1-25](#)  
regulated [3-1-17](#)

Contaminated clothing [B-1-4](#)

Control [4-1-1](#)  
treatment [3-1-20](#)

Control mechanism  
primary authorized [4-1-1](#)

Cooperative agreements [2-1-2](#)

Cooperators [2-1-2](#)

Corms [3-1-3](#)

Corn  
ear with shucks [3-1-21](#)  
unshucked ears [3-1-2](#)



---

Cornell University [2-1-2](#)  
Corns [3-1-21](#)  
Crates [3-1-3](#), [3-1-16](#)  
Crop rotation system [4-1-4](#)  
Crops  
  management sequence [4-1-4](#)  
Cysts [2-1-5](#), [2-3-12](#)

---

## D

Decomposed manure [3-1-2](#)  
Delimiting [2-2-3](#), [2-3-3](#)  
Detection [2-2-3](#), [2-3-3](#)  
Detection surveys  
  outside New York State [2-3-15](#)  
Diagram  
  example [A-1-19](#)  
  inspected property [A-1-21](#)  
Dry heat fumigation [3-1-23](#)  
Duties [2-3-6](#)

---

## E

Ear corn [3-1-21](#)  
Eggplants [2-1-5](#), [3-1-21](#), [3-1-24](#)  
Eggs [2-1-5](#)  
Eligibility  
  movement [3-1-24](#)  
  survey [2-3-14](#)

Emergency  
  action [B-1-4](#)  
  aid [B-1-1](#), [B-1-6](#)  
Equipment  
  mechanized soil moving [3-1-25](#)  
  regulated [3-1-17](#)  
Establishments [3-1-24](#)  
Exceptions [3-1-2](#)  
Export shipment [3-1-16](#)  
Exposed land [2-2-2](#)

---

## F

Fallow fields [3-1-27](#)  
Farm  
  equipment movement [3-1-25](#)  
  product containers [3-1-3](#)  
  tools [3-1-3](#)  
Farmer [4-1-4](#)  
Federal cooperators [2-1-3](#)  
Federal regulations [4-1-1](#)  
Female [2-1-5](#)  
Female nematode [2-1-5](#)  
Field  
  folder [2-4-28](#)  
  soil sampling [2-3-3](#)  
First aid rescue breathing [B-1-5](#)  
Forage crops [3-1-28](#)  
Fumigant  
  monitoring [B-1-3](#)  
  safety precautions [B-1-1](#)  
Fumigation  
  dry heat [3-1-23](#)



---

## G

Globodera rostochiensis (golden nematode) [2-1-1](#), [2-1-4](#)

Golden Nematode Quarantine 7CFR.85 [1-1-3](#)

Golf courses [3-1-27](#)

Grader stations [3-1-26](#)

Grader survey [2-3-11](#)

### Grain

fields [3-1-28](#)

small [3-1-22](#)

Grass sod [3-1-2](#)

Greenhouse [3-1-24](#)

Grid pattern [2-3-7](#)

Growers [2-2-4](#), [2-3-3](#)

### Guidelines

fumigant safety [B-1-2](#)

releasing land from regulation [3-1-27](#)

---

## H

Harvest [2-3-14](#)  
equipment [3-1-3](#)

### Hay

movement in approved containers [3-1-21](#)

regulated article [3-1-2](#)

Highways [3-1-27](#)

Home gardens [3-1-27](#)

Hosts [2-1-5](#)  
[3-1-25](#)

---

## I

Industrial areas [3-1-27](#)

### Infestation

areas [2-3-10](#)

property restrictions [4-1-4](#)

signs [2-1-7](#)

symptoms [2-3-14](#)

viable [2-3-14](#)

infestation [2-1-7](#)

### Inspection

grader station [3-1-26](#)

### Inspector

approved containers [3-1-16](#)

duties [2-3-6](#)

Irish potatoes [3-1-3](#)

---

## L

Labeling, bag [2-2-17](#)

### Laboratories

approved [3-1-24](#)

soil samples [3-1-2](#)

### Land

Category 3 Agricultural [3-1-27](#)

guidelines for releasing from regulation [3-1-27](#)

non-agricultural [3-1-27](#)

release from exposed status [2-3-14](#)

Larvae [2-1-5](#)

Limitations [3-1-22](#)

Lists [4-1-3](#)

Litter, plant [3-1-21](#)

Local cooperators [2-1-2](#)

---

## M

Mandatory surveys [2-3-12](#)

Manual samplers [2-3-3](#)

Manure, decomposed [3-1-2](#)

Maps [2-1-3](#), [2-4-28](#)

### Mechanical

(wheel) soil sampling [2-3-9](#)

cultivating equipment

[3-1-3](#)

equipment [2-3-9](#)

soil moving equipment

movement of used [3-1-25](#)

soil sampling equipment [2-2-18](#), [2-3-4](#)

Methyl bromide [3-1-19](#)

fumigant monitoring [B-1-3](#)

fumigation at NAP, chamber, or tarpaulin [3-1-26](#)

---

Mexico [3-1-4](#)

Modified 8-by-8 Block Method [2-3-8](#)

Monitoring fumigant [B-1-3](#)

Monoculture [2-1-5](#)

Movement  
of potatoes under limited permit (except seed potatoes) [3-1-20](#)  
regulated articles [3-1-18](#)  
soil to approved laboratory [3-1-24](#)  
soil to approved non-agricultural areas [3-1-23](#)  
to approved destinations [3-1-4](#)

Muck  
regulated article [3-1-2](#)

---

## N

Negative  
confirmed [3-1-19](#)  
survey [2-3-14](#)  
viability survey  
negative for viability [4-1-4](#)

New York  
guidelines for survey [2-3-2](#)  
infestation locations [2-1-4](#)

New York Certified Seed Potato Improvement Cooperative [2-1-2](#)

New York Department of Agriculture and Markets [2-1-2](#)  
agreement with [4-1-4](#)

Non-agricultural  
areas [3-1-23](#)  
land Category 1 [3-1-27](#)  
land Category 2 [3-1-27](#)

Non-chemical control [4-1-1](#)

---

Nurseries [3-1-28](#)

Nursery survey [2-3-11](#)

---

## O

Offshore States [3-1-4](#)

Original field folder [2-4-28](#)

Ornamental plants  
regulated article [3-1-3](#)  
tubers [3-1-21](#)

---

## P

Pallet boxes [3-1-16](#)

Peat [3-1-2](#)

Peripheral survey [2-3-11](#)

Permit  
determine if required [3-1-7](#)  
regulated articles [3-1-2](#)

Pest management program [4-1-4](#)

Pesticides  
authorized [3-1-19](#)

Phosphine [B-1-3](#)

Plant  
crowns [3-1-3](#)  
damage [2-1-6](#)  
litter [3-1-2, 3-1-21](#)

Plants  
ornamental [3-1-3](#)  
with roots [3-1-3, 3-1-21](#)



Poisoning signs and symptoms [B-1-6](#)

Positive find [2-3-12](#)

Possessions, U.S. [3-1-4](#)

Post resistant-variety treatment survey [2-3-13](#)

Post-crop survey [2-3-12](#)

Potatoes [2-1-5](#)  
 from noninfested fields [3-1-20](#)  
 production areas [2-3-14](#)  
 resistant varieties [4-1-3](#)

Potting soil  
 movement [3-1-23](#)  
 steam fumigation [3-1-24](#)

PPQ Form 312, Golden Nematode Survey [2-4-28](#),  
[A-1-17](#), [A-1-24](#)  
 example [A-1-17](#)  
 instructions [A-1-21](#)

PPQ Form 391, Specimens for  
 Determination [A-1-25](#)  
 distribution [A-1-28](#)  
 field folder [2-4-28](#)

PPQ Form 519, Compliance Agreement [A-1-29](#)  
 distribution [A-1-34](#)  
 example [A-1-29](#)  
 instructions [A-1-33](#)  
 purpose [A-1-33](#)

PPQ Officers [3-1-18](#)

Probes [2-3-9](#)

Program pest management [4-1-4](#)

Property [4-1-4](#)

---

## Q

Quarantine areas [2-1-3](#)

---

## R

Race  
 Ro1 [2-1-4](#), [4-1-3](#), [D-1-2](#)

Racetracks [3-1-27](#)

Records  
 bag labeling [2-3-15](#)

Recreational land [3-1-27](#)

Regulated articles [3-1-2](#), [3-1-18](#)

Regulated land [2-2-2](#)

Regulation [3-1-27](#)

Residential areas [3-1-27](#)

Resistant potato varieties [4-1-3](#)

Resistant varieties [2-3-13](#), [3-1-20](#), [4-1-1](#)

Rested field survey [2-3-12](#)

Restrictions [4-1-4](#)

Rhizomes [3-1-3](#), [3-1-21](#)

Riding academies [3-1-27](#)

Ro1 [2-1-4](#), [4-1-3](#)

Root crops [3-1-3](#), [3-1-21](#)

Roots [3-1-3](#)

Routine soil survey [2-3-2](#)

---

## S

Safety [B-1-1](#)

Sample bags [2-3-7](#)

Sampling  
 8 x 8 block method [2-3-7](#)  
 peripheral survey [2-3-11](#)  
 simplified 8 x 8 block method [2-3-8](#)

Sanitation [2-2-2](#), [2-2-18](#), [3-1-19](#)

Seed potato fields [2-2-19](#), [2-3-5](#)

Selected area soil sampling [2-3-10](#)

Selection guide [2-3-10](#)

Shipment, offshore [3-1-4](#)

Shippers [3-1-18](#)

Sketch [A-1-21](#)

Small grains [3-1-3](#), [3-1-22](#)

Sod [3-1-2](#), [3-1-21](#)  
 farms [3-1-28](#)

Soil  
 dry heat fumigation [3-1-23](#)  
 limitations for fumigation [3-1-22](#)  
 movement to approved laboratory [3-1-24](#)  
 movement to approved non-agricultural areas [3-1-23](#)  
 regulated article [3-1-2](#)  
 regulated samples [3-1-2](#)  
 removing compacted [3-1-25](#)  
 sample movement [3-1-23](#)  
 sampling  
   mechanical (wheel) [2-3-9](#)  
 selected area sampling [2-3-10](#)  
 selection guide [2-3-10](#)  
 surveys [2-3-3](#)  
 temperature [2-1-5](#)

Soil-free medium [3-1-25](#)

Soybeans  
 other than for seed [3-1-22](#)  
 regulated article [3-1-3](#)

Special instructions [3-1-4](#)

State cooperators [2-1-2](#)

State regulations [4-1-1](#)

Steam cleaning [2-2-18](#), [2-2-19](#), [2-3-4](#), [2-3-5](#), [3-1-25](#)

Steam fumigation [3-1-24](#)

Steam heat treatment [4-1-1](#)

Straw [3-1-2](#), [3-1-21](#)

Sugar beets [3-1-22](#)

Sulfuryl fluoride [B-1-3](#)

Surveillance survey [2-3-14](#)

Surveys [4-1-1](#)  
 biometrics [2-3-14](#)  
 confirmation [2-3-12](#)  
 mandatory [2-3-12](#)  
 negative [2-3-14](#)  
 Nursery [2-3-11](#)  
 outside New York State [2-3-15](#)  
 outside regulated areas [2-3-14](#)  
 peripheral [2-3-11](#)  
 post resistant variety [2-3-13](#)  
 post-crop [2-3-12](#)  
 release land from exposed status [2-3-14](#)  
 rested field [2-3-12](#)  
 routine soil [2-3-2](#)  
 surveillance [2-3-14](#)  
 suspect area [2-3-14](#)  
 symptom [2-3-14](#)

Suspect area survey [2-3-14](#)

Symptom survey [2-3-14](#)

Systematic  
 manual soil sampling [2-3-6](#)  
 use [4-1-1](#)

---

## T

Territories [3-1-4](#)

Tomatoes [2-1-5](#), [3-1-21](#)  
 product areas [2-3-14](#)  
 transplants [3-1-24](#)

Transplants [3-1-24](#)

Treatments  
 Irish Potatoes (except seed potatoes) [3-1-20](#)

Trucks [3-1-16](#), [3-1-28](#)

Tubers [3-1-3](#)

---

## U

Update record  
 for the APM [1-1-ii](#)

USDA-ARS [2-1-2](#)

---

## V

Vehicles [2-2-18](#), [2-3-4](#)

Viability survey [4-1-4](#)

Viable cyst [2-3-12](#)

Vikane [B-1-3](#)

---

## W

Water-under-pressure (pressure wash) [3-1-25](#)

Wheel (mechanical) soil sampling [2-3-9](#)