

2013 *Phytophthora ramorum* Nursery Survey



Revised: March 2013

United States Department of Agriculture Animal and Plant Health Inspection Service Plant Protection and Quarantine

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APPENDICES

Biology of *Phytophthora ramorum* APHIS List of Regulated Hosts and Plants Associated with *P. ramorum* Host Genera in U.S. Nurseries Symptoms Associated with *P. ramorum* Suggested Supplies and Equipment Nursery Survey Data Collection Form Water Sampling Protocol

NURSERY SURVEY OBJECTIVES

The objective of the *Phytophthora ramorum* Nursery Survey is to detect the presence of *P. ramorum* in nurseries in the United States. This objective will be accomplished by surveying nurseries at risk of harboring or distributing *P. ramorum* infected plants.

BIOLOGY OF PHYTOPHTHORA RAMORUM

Please read the appendix on the Biology of *Phytophthora ramorum* before starting surveys.

DEFINITIONS

Compost pile:	A heap of decaying organic matter layered and mixed together and allowed to compost; used to improve soil structure and provide nutrients (components include plant parts, manure, etc.).
Confirmed Positive:	The presence of <i>P. ramorum</i> is confirmed in a diagnostic test conducted by analysts/labs with federal confirmatory authority by utilizing the APHIS <i>P. ramorum</i> Diagnostic Work Instructions. This final determination of a positive sample allows for federal regulatory action. Report the positive to the APHIS PPQ Operations Manager w/in 24 hours.
Cull Pile:	An area where discarded plant material, container mix (growing media), pots are deposited; also known as a waste or trash pile.
Final Determination:	The last diagnostic result(s) necessary for a regulatory sample indicating whether <i>Phytophthora ramorum</i> is present or not. The final determination test(s) are conducted by analysts/labs with federal confirmatory authority. If the final determination is positive, report to the APHIS PPQ Operations Manager w/in 24 hours. Regulatory action may commence on this positive. <i>See</i> confirmed positive.
НАР:	Host and associated host plants listed on the official APHIS List of Regulated Hosts and Plants Associated with <i>Phytophthora ramorum</i> . http://www.aphis.usda.gov/plant_health/plant_pest_info/pram/downl oads/pdf_files/usdaprlist.pdf
High Risk Genera: 0	Camellia, Rhododendron, Pieris, Viburnum and Kalmia.
Infected plants:	Plants officially confirmed as being infected with <i>P. ramorum</i> , based on the use of APHIS approved diagnostics, and following the PASS system.
Non-PASS:	Once a nursery is Confirmed Positive by APHIS during a given calendar year, all subsequent samples are considered "Non-PASS", meaning a NPPLAP accredited lab outside of APHIS can make the final determination utilizing APHIS NPPLAP diagnostic work instructions. If the diagnostic result is positive, the sample is Confirmed Positive and reported to the APHIS PPQ Operations Manager w/in 24 hours. Regulatory action can commence based on that positive sample.
Nursery:	Any location where nursery stock is grown, propagated, stored, or sold, or any location from which nursery stock is distributed.

Nursery block:	A contiguous grouping of plants separated by some distance by a path, preferably at least two meters, from other contiguous groupings of plants.
Nursery Dealer:	Nurseries that are resellers – wholesale or retail – of nursery plants.
Nursery Grower:	Nurseries that grow nursery stock; synonymous with propagator.
Nursery stock:	Any plants for planting, including houseplants, propagative material that are grown in a nursery and tree seedlings for reforestation. In this and other protocols, the terms nursery stock and "plant" are often used synonymously, but not exclusively.
PASS:	Potential Actionable Suspect Sample: it is the initial sample Confirmed Positive by APHIS for a given nursery in the calendar year. More specifically, when a NPPLAP accredited lab detects <i>P. ramorum</i> in a sample of a nursery not yet confirmed positive by APHIS during a given calendar year, it is consider "PASS" and must be routed to APHIS for final determination .
Sample:	For <i>P. ramorum</i> plant sampling, a sample refers to a single bag of leaves. Each sample is to contain a minimum of 5 symptomatic (unhealthy) leaves from large-leaf species; 5-12 leaves if possible, and, for small leaf species, include the terminal 3 inches of a symptomatic branch. Regulatory action may commence on one sample confirmed positive . In <i>P. ramorum</i> diagnostics, sample may refer to a 25 microliter aliquot of DNA or a culture derived from one plant sample.
Soil:	The loose surface material of the earth in most cases consisting of disintegrated rock with an admixture of organic material. The reference to soil in this protocol is the surface or substrate under plant containers, the bare ground and /or gravel; often with plant debris, peat and bark fines washed from plant containers. Substrates in the field on which potted plants are located; often this consists of peat and bark fines washed from pots, plant debris, soil, gravel, or any combination of these.

TIMING NURSERY SURVEYS

Nurseries should be surveyed at a time when climatic conditions conducive to *P. ramorum* disease expression have occurred, providing the <u>best opportunity for expression of</u> <u>symptoms due to *P. ramorum*.</u>

- Research suggests that the most favorable climate for the expression of symptoms includes ambient temperatures between 3°C and 28°C (optimum 20°C) and free moisture present on host tissue for at least 12 hours over 10 or more days. In many areas this is primarily in the spring and may occur in the fall.
- Greenhouses, hot houses and nursery beds under shade cloth or overhead irrigation should be considered micro-climates where optimum conditions are governed more by nursery and market practices than external conditions. These micro-climates may provide suitable conditions for *P. ramorum* expression at times of the year which would not be considered ideal based on local environmental conditions. These areas must be taken into consideration when selecting sites and determining times to survey.
- Disease expression typically begins between 30 and 90 days after bud-break. In many locations, the survey should begin after the spring flush is underway, when some of the leaves have fully expanded. Plants can express symptoms throughout the growing season, though isolation of the pathogen from diseased tissue may be more difficult during hot and dry periods.
- It is also important to consider the times when nurseries are scheduled to receive shipments in the spring and fall in order to plan surveys when a majority of host plants will be present during conducive climate conditions for *P. ramorum*.

RISK FACTORS AND SELECTING NURSERIES TO SURVEY

For the Farm Bill <u>P. ramorum Program Directed Nursery Survey</u> or CAPs bundled nursery surveys that include *P. ramorum* (i.e., any survey funded by APHIS via Farm Bill or program dollars), the criteria for nursery selection is outlined below. If you have questions, contact your State Plant Health Director or the *P. ramorum* Field Operations Manager. The *P. ramorum* survey aims to fill in gaps in our knowledge about the distribution of the pathogen across the U.S. with the ultimate goal of protecting the biodiversity and natural resources of states with high risk environments. Risk factors include both the risk to disseminate *P. ramorum* and to receive *P. ramorum*.

Nursery Risk Factors

- Nurseries that propagate HAP, particularly the high risk genera (*Camellia*, *Rhododendron*, *Viburnum*, *Pieris*, *Kalmia*) and sell them to residents or nurseries in high risk environments, or to other wholesale nurseries
- Nurseries that ship HAP interstate wholesale or retail to citizens
- Nurseries that receive host plants from known infected suppliers (i.e., trace forward nurseries)
- Nurseries previously positive for *P. ramorum*

Nursery Selection Priority

- In states containing areas considered at-risk for disease outbreak if *P. ramorum* were introduced, surveys should target propagation, wholesale, broker, and retail nurseries containing *P. ramorum* host plants. (As certain geographical regions of the U.S. could be at risk to disease outbreak, such as forests, parks, or greenbelts containing multiple stories of host genera, see contact list at the bottom of this document for the latest risk assessment information.)
- In areas considered high risk to disease outbreak, include survey of the environment adjacent (within 10 meters) to nurseries with high risk genera or which have been positive for *P. ramorum* (in plants, soil, water and streams).
- Nurseries in states or locations not considered at-risk, but which sell host plants to residents or other businesses (landscapers, brokers, other nurseries) in high risk environments should also be surveyed.
- In states considered to have low or no risk environs, give priority to nurseries that ship *P. ramorum* host plants interstate, particularly the high risk genera (*Camellia, Rhododendron, Viburnum, Pieris, Kalmia*); ensure a portion of your nursery selection includes these inter-state shippers (propagators, wholesalers, or retailers). Next in priority are intrastate HAP propagators, intrastate HAP wholesalers, and then intrastate HAP retailers.
- Nurseries that received potentially infected stock in the previous year.
- For states conducting surveys for multiple years, rotate your priority nurseries to ensure coverage of all nurseries with some risk factor:
 - Previously infested
 - Received host plants from known infected suppliers (i.e., trace forward nurseries).
 - Never been inspected
 - Not inspected during the previous year
 - Large inventory of HAP (particularly those in the five genera believed to be most susceptible to *P. ramorum* infection in the nursery settings: *Camellia, Rhododendron, Viburnum, Pieris,* and *Kalmia*)
 - Both interstate and intrastate businesses
 - Balance your selection between nursery growers (production/propagative nurseries) and nursery wholesalers, retailers, brokers, and dealers.

Plant Selection Priority

Inspect plants on the official <u>APHIS List of Regulated Hosts and Plants Associated with</u> <u>Phytophthora ramorum</u>. The complete and up-to-date host list is available at the APHIS *P*. *ramorum* Web site. You should consult it for the latest list of plants before beginning your inspections. There are five plant genera most frequently reported to be infected by *P. ramorum* in U.S. nursery settings. These hosts are considered "high risk"; be sure to inspect these five genera while surveying HAP in the nursery. The high risk genera are:

- Camellia
- Rhododendron
- Viburnum
- Pieris
- Kalmia

PLANT SYMPTOMS AND SAMPLING FOR P. RAMORUM

Plant Symptom Resources

Inspectors should receive training in identifying symptoms associated with *P. ramorum* on host plants. At a minimum they should review photographs of the wide range of symptoms possible before starting the survey. Photographs of typical and atypical symptoms are available in Appendix D.

Remember that symptoms of *P. ramorum* are often not "typical" and over reliance on identification by these symptoms could result in infected plants remaining undetected – the greatest chance of detecting *P. ramorum* infections is though the collection of ANY unhealthy looking plant tissue for laboratory analysis.

Photos of *P. ramorum* symptoms are available at the following websites: APHIS PPQ *P. ramorum* Program; the USDA *Phytophthora ramorum* Educate to Detect (PRED) Program; California Oak Mortality Task Force (COMTF); University of California at Davis Nursery Guide for Diseases of *Phytophthora ramorum* on Ornamentals: Diagnosis and Management; and the OSU *P. ramorum* biology, symptoms, and diagnosis module of their online training:

- APHIS: <u>http://www.aphis.usda.gov/plant_health/plant_pest_info/pram</u>
- PRED: <u>http://www.ncpmc.org/alerts/suddenoakdeath/pred.cfm</u>
- COMTF: <u>http://www.suddenoakdeath.org</u>
- UC Davis: <u>http://anrcatalog.ucdavis.edu/pdf/8156.pdf</u>
- OSU: <u>http://oregonstate.edu/instruct/dce/phytophthora/module1.html</u>

Sampling Principles

There are two basic principles that should govern the inspection and sampling processes.

- 1. *P. ramorum* cannot be diagnosed by a visual inspection of symptoms alone, only laboratory testing can provide a definitive diagnosis.
- 2. If there is any doubt as to whether the symptoms observed could be caused by *P*. *ramorum*, collect a sample.

Plants chosen to be inspected should be carefully scrutinized. Foliar symptoms of *P. ramorum* infection are highly variable (see Appendix D) and can range from pinpoint discolorations on the leaf surface to large "V" shaped lesions along the leaf mid-vein. Include inspection of the lower portions of plants where conditions favoring *P. ramorum* would be present. Moisture will tend to be present for longer periods of time on the plant surfaces on the lower portions of the plants also resulting in higher humidity. Shading on the lower portions of the plants can promote cooler temperatures and offer protection from the effects of UV rays on spores. Pay attention to leaf areas where water would run off or persist the longest such as the midrib and leaf tips. In some hosts (*Camellia & Rhododendron*) low rates of infection can cause premature leaf drop, yielding infected plants that appear to be asymptomatic. As a result, leaves found in the pot or on the ground below the plant should also be checked for possible symptoms and collected for laboratory analysis.

Collect samples of **any and all** plant tissue that appears unhealthy. For *P. ramorum* plant sampling, a **sample** refers to a <u>single bag of leaves</u>. Each sample is to contain a minimum of 5

symptomatic (unhealthy) leaves from large-leaf species; 5-12 leaves if possible, and, for small leaf species, include the terminal 3 inches of a symptomatic branch with the leaf samples. Examine the leaves carefully; chances are much smaller lesions will be present on other leaves of the same plant. It is strongly encouraged that each sample is from one plant, however, if there are not enough symptomatic leaves on the one plant, collect symptomatic leaves (if present) from other plants in the same block (i.e. at the lowest taxonomic level possible, plants of the same cultivar/variety is preferred, then species, then genus). The situation may occur where only one plant with few symptomatic leaves may be present representing a block. In this case, sample the symptomatic leaves and adjacent leaves for the sample. If there is a large amount of unhealthy tissue, collect as many samples as needed to fully <u>represent</u> the symptomas seen on a genus/species/variety/block basis. This does not mean sampling every single symptomatic plant, but sampling enough of them in any given block so that you are sure to give the lab the material it needs to make a correct diagnosis. It is important to establish good communication and feedback with the lab which the samples are being sent to, in order to continue to optimize the collection of sample material in the field.

Do not be intimidated by a lack of certainty as to what *P. ramorum* symptoms might look like. Remember, other common *Phytophthoras*, other pathogens and environmental stressors can cause similar symptoms that cannot be identified based on visual inspection. Do <u>not</u> collect samples from healthy, asymptomatic plants. If no unhealthy plants are observed, note how many healthy HAP plants were inspected.

Sampling by Symptom Type

Leaf Spots and Lesions

- Collect symptomatic leaves.
 - Symptomatic fallen leaves *within the pot* of a symptomatic plant can be included in the sample provided they are not exhibiting extensive decay.
 - For plants with very small leaves or needles, samples can be submitted as twig sections with the leaves attached. In these cases try to ensure that the sample has a total of approx. a 3" x 3" leaf surface area.

Twig Dieback

- Cut the twigs below the cankered regions (well into healthy tissue).
- Sterilize pruning equipment between samples using a dilute (10%) bleach solution or a quaternary ammonium solution

Cankers on Boles and Branches of Trees

- Follow procedures in your state for surveying and sampling trees.
- In some states nursery inspectors may sample trees while in other states forestry or other officials may be asked to sample trees.

PREPARING FOR NURSERY SURVEYS

Supplies and Equipment Check List

- <u>APHIS List of Regulated Hosts and Plants Proven or Associated with Phytophthora</u> <u>ramorum</u> (January 2012): <u>http://www.aphis.usda.gov/plant_health/plant_pest_info/pram/</u>
- Nursery maps and nursery inventory
- Clipboard, PPQ <u>391 forms</u>, nursery survey forms, PDA, paper, etc.
- Camera
- Writing pen
- Permanent marking pen
- Latex gloves (new gloves or spray/wash gloves for each new sample)
- Snips to sample twigs and branches
- GPS
- Half-apron with front pockets to carry supplies
- Spray bottle of an approved disinfectant for *P. ramorum*
- Re-sealable plastic sample bags
- Bigger collection bag to carry samples in while surveying
- Cooler, coolant, and newspapers to keep samples cool until mailed
- Larger bags for mailing samples (must arrive in lab double-bagged)
- Box for mailing samples
- Flagging, pin flags, or label sticks to mark sampled plants/blocks

SURVEYING THE NURSERY AND COLLECTING SAMPLES

Surveying

1) If available, obtain and review an inventory and a location map of host plants in the nursery to help determine where the plants to be inspected are located. Include areas of cull piles containing host material, 'plant hospitals' or low vigor plants reduced for sale, and areas which may include host plant returns.

2) Begin the survey by conducting a visual assessment of the nursery as a whole. During this survey identify any low lying areas, standing water, the nursery layout, source of water, the general condition of the plants and nursery environment. Use this information to help guide your plant inspection and sampling. For *P. ramorum* nursery surveys funded by Farm Bill, it is a requirement that the information within the <u>nursery survey data collection form</u> is collected in the nursery and entered into <u>IPHIS</u>. Data entry into NAPIS is not required for the *P. ramorum* program. *See* more information on the Farm Bill funded P. *ramorum* Program Directed Nursery Survey requirements:

http://www.aphis.usda.gov/plant_health/plant_pest_info/pram/survey.shtml

Collecting Samples

3) Samples collected from this and other APHIS funded surveys that are suspect positive must be forwarded to APHIS (or a NPPLAP accredited lab) for further testing. Samples confirmed positive must be reported to the SPRO/SPHD, then to the P. ramorum Operations Manager within 24 hours of the SPRO/SPHD notification.

4) Follow decontamination procedures of personnel, tools, and equipment between blocks in the nursery, between host genera within a nursery, and between nurseries. Decontaminate all equipment you use to take samples between blocks of nursery stock and before leaving a nursery. Use a spray bottle containing a dilute (10%) bleach solution or a quaternary ammonium solution to treat all tools between nursery blocks. Brush loose dirt from boots and shoes and then spray boots or shoes with disinfection solution in spray bottle between nursery blocks.

5) Determining the Number of Plants to Inspect

Visually inspect a minimum of number of host plants in each nursery at random based on Table 1. At the discretion of the inspector, more plants may be visually inspected and sampled if conditions suggest this is needed.

Host plants in the five highly susceptible HAP genera listed previously should be inspected, by genus, at the rates listed in the table. All other host plants are inspected, as a whole, at the rate specified in the table.

For example, if a nursery has 3,000 Camellias, 6000 Rhododendron and 8000 plants in host genera other than the six highly susceptible genera minimally inspect 1,055 Camellias, 1,087 Rhododendrons and 1,087 randomly selected other HAP plants.

nursery.	
Host and Associated Plants	95% Confidence Limit of
Per Nursery	Detecting 0.5% Disease
n<500	All plants
501 <n<1,000< td=""><td>842</td></n<1,000<>	842
1001 <n<5,000< td=""><td>1055</td></n<5,000<>	1055
5,001 <n<10,000< td=""><td>1087</td></n<10,000<>	1087
n>10,001	1115

Table 1 *Determining the number of hosts and associated plants for visual inspection within a nursery.

*Numbers are the minimum number of host and associated host plants that must be inspected in a nursery to ensure detection at a 95% confidence level when the disease is present in 0.5% of the plants, when 75% of infected plants express symptoms.

6) A sample is a <u>single bag of leaves</u> that is from a single plant, or, if from more than one plant, is from plants of the same cultivar and definitely from within the same block (for example, the sample contains leaves from several plants of *Rhododendron* 'Roseum Pink'). If necessary, the sample can be a composite from the same genus/species of multiple cultivars as long as they are from the same block (for example, all of the leaves in a sample are from "*Kalmia latifolia*". If at all possible, avoid samples collected and labeled at the genus level (e.g., *Camellia* sp.); this provides very little epidemiological assistance if there is a positive.

7) Using a permanent marker, label the sample bag: time, date, sample number, **genus species cultivar**, collector's identification number, location of sample collection, etc. Mark the sampled plant with flagging tape, stake, etc., and label with the corresponding sample number. This will facilitate any additional work in the event of a positive or the need for a second sample.

8) Do not add extra moisture to the sample to keep it fresh. Do not wrap leaves in paper towel when shipping. The extra moisture/paper towels speed deterioration of the sample. Remove gloves and place sample bag in a second protective bag. >> See full checklist on collecting: "Sample Handling and Submission Protocol" below.

9) Sample the water in any holding pond(s), ditches, creeks, drainage pipes, drainage systems or standing water. *See* Water Sampling Protocol *Updated February 2013*

10) Surveyors may wish to draw a map of the nursery indicating areas inspected and sampled, in addition to flagging or marking plants/blocks sampled. This can be very useful if resampling is necessary.

11) Identifying Other Areas of the Nursery to Inspect: Areas of 'Sale Plants' or returns: Sample if symptomatic plant tissues are observed. Cull and/or compost piles: Locate and inspect cull piles of plant materials that have been discarded or taken off sale. Sample if symptomatic plant tissues are observed. Inspect these piles after you have completed inspections of the rest of the nursery.

SAMPLE HANDLING AND SUBMISSION PROTOCOL

- Always write out the identifying label remarks on the outside of the bag with a permanent marker
 - Attach labels on the outside of bags since labels inside the bag may deteriorate due to moisture and become illegible
 - Include on all labels with a permanent marker: time, date, collector's identification number, location of sample site, sample number
- **Do not** add extra moisture to the sample to keep it fresh. <u>Do not</u> wrap leaves in paper towel when shipping. The extra moisture/paper towels speed deterioration of the sample. Remove gloves and place sample bag in a second protective bag.
- To provide extra insurance against accidental release during shipping, the labeled specimen bags should be double-bagged i.e. first place the specimen in a self-locking labeled plastic bag and then place that labeled specimen bag(s) within a second self-locking plastic bag. **The Form 391 should be placed inside the outer bag**
- Samples should be placed in a cooler out of the sun as soon as possible. When sampling large areas, coolers should be brought out to the sampling areas. Samples can be exposed to high temperatures quickly when placed in plastic bags in sunlight for even short periods of time. If it is not possible to have coolers in the area of sampling or due to lack of coolers, place the samples in a shaded area until they can be collected and placed in a cooler as soon as possible.

- Refrigerate samples while awaiting shipment to the diagnostic laboratory. Fresh samples produce a higher quality diagnostic result. If possible, ship with an ice pack. Please do not ship on Fridays. It is better to hold them in the refrigerator over the weekend than to have them sit on a truck or warehouse dock in undesirable environmental conditions. Mail or deliver the sample as soon as possible to preserve freshness (if mailing use overnight mail).
- Place double-bagged samples in a sturdy cardboard box or heavy Styrofoam container so that the samples are not damaged during shipping and handling. Thoroughly seal all seams on the container with shipping tape.

Notifying the Lab

Submitters should notify the lab when surveys are planned in order to allow the lab time to ensure that necessary supplies are available and they are prepared to receive samples for processing. This will provide the lab with the best opportunity to process the samples as they are received rather than the possibility of losing important samples or the necessity of resurveying due to loss or deterioration of samples. Submitters should contact the lab prior to sending the samples (see contact list below). Have ready the required information: 1) tracking number, 2) number of samples being shipped, 3) the disease being tested for. All samples must have either a completed PPQ Form 391 or equivalent documentation. The lab may be your cooperating NPDN lab, or other cooperating NPPLAP accredited lab, or an APHIS PPQ lab (see Contact List at the end of this document). If submitters aren't sure which lab to send samples to, contact the PPQ NIS Domestic Diagnostics Coordinator, Joel.P.Floyd@aphis.usda.gov (301) 851-2115) or the PPQ *P. ramorum* Operations Manager, Stacy.E.Scott@aphis.usda.gov (970) 494-7577.

Sample Submission Information

For all samples, particularly any that are ELISA or ImmunoStrip positive for *Phytophthora* sp., complete a PPQ Form 391 to accompany the sample. If the PPQ391's are electronic, they can be emailed when notifying the lab about the pending shipment, but it is always good practice to attach a hardcopy in the sample. **Fill out blocks 1-5, 7, 10, 11, 16, 22, 23** (see green circled items below) in the <u>PPQ Form 391</u> instruction sheet.

Instructions

Use PPQ Form 391, Specimens for Determination, for domestic collections (warehouse inspections, local and individual collecting, special survey programs, export certification).

BLOCK	INSTRUCTIONS					
	 Assign a number for each collection beginning the year, followed by the collector's initials and collector's number 					
1	EXAMPLE In 2001, Brian K. Long collected his first specimen for determination of the year. His first collection number is 01-BLK-001					
	2. Enter the collection number					
2	Enter date					
3	Check block to indicate Agency submitting specimens for identification					
4	Enter name of sender					
5	Enter type of property specimen obtained from (farm, nursery, feedmill, etc.)					
6	Enter address					
7/	Enter name and address of property owner					
8A-8L	Check all appropriate blocks					
9	Leave Blank					
10	Enter scientific name of host, Genus Species, particularly Cultvar name					
11	Enter quantity of host and plants affected					
12	Check block to indicate distribution of plant					
13	Check appropriate blocks to indicate plant parts affected					
14	Check block to indicate pest distribution					
15	Check appropriate block to indicate type of specimen Enter number specimens submitted under appropriate column					
(16)	Enter sampling method					
17	Enter type of trap and lure					
18	Enter trap number					
18 19	Enter trap number Enter X in block to indicate isolated or general plant symptoms					
18 19 20	Enter trap number Enter X in block to indicate isolated or general plant symptoms Enter X in appropriate block for weed density					
18 19 20 21	Enter trap number Enter X in block to indicate isolated or general plant symptoms Enter X in appropriate block for weed density Enter X in appropriate block for weed growth stage					
18 19 20 21 22	Enter trap number Enter X in block to indicate isolated or general plant symptoms Enter X in appropriate block for weed density Enter X in appropriate block for weed growth stage Provide a brief explanation if Prompt or URGENT identification is requested					
18 19 20 21 22 23	Enter trap number Enter X in block to indicate isolated or general plant symptoms Enter X in appropriate block for weed density Enter X in appropriate block for weed growth stage Provide a brief explanation if Prompt or URGENT identification is requested Enter a tentative determination if you made one					

Distribution of PPQ Form 391

Distribute PPQ Form 391 as follows:

- Send Original along with the sample to your Area Identifier.
 Retain and file a copy for your records.

REPORTING RESULTS

Survey Accomplishment Reporting Track the following data for *P. ramorum* Nursery Surveys funded by APHIS PPQ via program or Farm Bill dollars:

State	Budget (actual)	Number of Nurseries Surveyed in CY12/13/14	Number of water samples tested	Number of water samples positive	Number of soil samples tested	Number of soil samples positive	Number of plant samples tested	Number of plant samples positive
		CY12						
		CY13						
		CY14						

These data are critical to APHIS as we account for money spent on specific goals and objectives. Please keep this simple tracking table up to date as the survey season progresses. Note: track survey accomplishments in the <u>calendar year</u> they occurred regardless of which fiscal year they were funded.

Sample Diagnostics Reporting

For the *Phytophthora ramorum* program, any sample collected within a nursery that is ELISA or ImmunoStrip positive for the genus *Phytophthora*, requires further diagnostics to determine if the species is *P. ramorum*. These samples must be forwarded to your cooperating <u>NPPLAP</u> accredited lab for further diagnostics. Samples **confirmed positive** must be reported to the SPRO/SPHD, then to the *P. ramorum* Operations Manager within 24 hours of the SPRO/SPHD notification.

POINTS OF CONTACT - APHIS PPQ P. RAMORUM NURSERY SURVEY

Your State Plant Health Director Pest Survey Specialist State Survey Coordinator

Stacy Scott Field Operations Manager for *P. ramorum* (970) 494-7577 <u>Stacy.e.Scott@aphis.usda.gov</u>

Dr. Prakash Hebbar Policy Manager for *P. ramorum* (301) 851-2228 Prakash.Hebbar@aphis.usda.gov

Joel Floyd PPQ NIS Domestic Diagnostics Coordinator (301) 851-2115) Joel.P.Floyd@aphis.usda.gov IPHIS Data Input: Dave Kowalski Data Manager, Data Analysis Risk and Targeting, USDA/APHIS/PPQ 970-494-7510 (cell 970-214-2729) david.g.kowalski@aphis.usda.gov

<u>NPPLAP Accredited Labs</u> *See* Diagnostic Section on PPQ *P. ramorum* Webpage: <u>http://www.aphis.usda.gov/plant_health/plant_pest_info/pram/protocols.shtml#diag</u>

PPQ Domestic Diagnostic Labs				
Grace O'Keefe	Dr. Craig Webb	Sample Diagnostics		
USDA APHIS PPQ	USDA APHIS PPQ	USDA-APHIS-PPQ-CPHST		
Diagnostic Laboratory	Diagnostic Laboratory	BARC-East, Bldg. 580		
Pennsylvania State University	Kansas State University	Powder Mill Road		
105 Buckhout Lab	4024 Throckmorton Plant	Beltsville, MD 20705-2350		
University Park, PA 16802	Sciences Center	(301)313-9204		
(814)865-9896	Manhattan, KS 66506	<u>APHIS-</u>		
grace.okeefe@aphis.usda.gov	(785) 532-1349	PPQCPHSTBeltsvilleSampleDiagnostics@aphis.usda.gov		
	craig.a.webb@aphis.usda.gov			

<u>FRONT</u>