

Phytoplasma Sample Routing for CAPS and Farm Bill

Last updated: 4/17/17

Samples:

All symptomatic suspect phytoplasma samples should be received double bagged and include a [PPQ 391 Form](#) (fillable and available on the APHIS website). Each sample from different plants and localities (*i.e.*, each plant sample) should have its own PPQ Form 391 per instructions in the [Phytoplasma Sample Submission](#) document. Each screening lab is to be notified prior to any samples being sent.

Laboratory Analysis:

Each sample should be screened using the DNA extraction and qPCR work instructions used in the training session by CPHST Beltsville. For up-to-date work instructions or any issues with the protocols contact Stefano Costanzo (301-313-9268; Stefano.Costanzo@aphis.usda.gov).

Negative Results

Negative results can be communicated by e-mail back to the submitter with Steve Bullington, USDA-APHIS-PPQ Domestic Diagnostics Coordinator, copied on the message. Stephen.W.Bullington@aphis.usda.gov

Positive Results:

All non-palm (*e.g.*, apple, grape, stone fruit, and pine) phytoplasma positive DNA samples should be sent to Dr. Robert Davis, with the exception of phytoplasma positive fruit trees or grapevines from Pennsylvania. The X-disease phytoplasma group (16SrIII) is common in Pennsylvania, and this state has its own process for routing and submitting 16SrIII phytoplasma positives. The state will continue to forward finds on new host plants or samples that are not straightforward in their identification.

Dr. Brian Bahder should receive all palm phytoplasma positive DNA samples with the exception of palm phytoplasma samples collected from Texas and Florida with the authorization of the [State Plant Regulatory Official \(SPRO\)](#) of the state of origin. Since palm phytoplasmas (Group 16SrIV) are known to occur in Texas (16SrIV-D) and Florida (16SrIV-A, D, and F), these states have their own process for routing and reporting 16SrIV phytoplasma positives.

The DNA should be labeled exactly the same as the leaf/tissue sample from which the DNA was extracted. The PPQ form 391 should also be sent with the sample.

Dr. Robert Davis

USDA-Agricultural Research Service
Molecular Plant Pathology Laboratory
Bldg 004, Room 220 / 221
10300 Baltimore Avenue
Beltsville, MD, 20705

Voice: 301-504-5745 or -6290
Fax: 301-504-5449
Email: robert.davis@ars.usda.gov

Dr. Brian Bahder

University of Florida
Entomology and Nematology Dept.,
FLREC 3205 College Avenue
Fort Lauderdale, FL 33314, USA
Voice: 954-577-6300
Email: bbahder@ufl.edu

If the sample is found to be positive for a phytoplasma, Dr. Davis and Dr. Bahder will notify Dr. Mark Nakhla at the PPQ CPHST Beltsville laboratory by email with a PDF attachment of the completed PPQ form 391, with results indicated in block #21 with a narrative of the tests performed.

Sample Diagnostics
USDA-APHIS-PPQ-CPHST
BARC-East, Bldg. 580 Powder Mill Road
Beltsville, MD 20705-2350
Phone: (301) 504-7100, VOIP: (301) 313-9200
Group E-mail Address:
APHIS-PPQCPHSTBeltsvilleSampleDiagnostics@aphis.usda.gov

If the phytoplasma is exotic to the United States, the PPQ CPHST Beltsville lab will obtain an official plant/tissue sample for confirmatory testing. The CPHST Beltsville Lab has the necessary permits to receive the infected material sent overnight in a properly secured crushproof container with the completed PPQ form 391. An e-mail notification with the overnight carrier tracking number needs to be sent to:

APHIS-PPQCPHSTBeltsvilleSampleDiagnostics@aphis.usda.gov.

For Pennsylvania phytoplasmas that are new state or host records, or palm phytoplasmas that are new state records only and not exotic to the United States, Steve Bullington and Mark Nakhla should be notified of the confirmation so that the state of origin can be notified via official results communications protocols.

PPQ Form 391

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to, a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 0579-0377. The time required to complete this information collection is estimated to average .25 hours per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information.					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.					OMB APPROVED 0579-0010 EXP. DATE 02/2017			
UNITED STATES DEPARTMENT OF AGRICULTURE ANIMAL AND PLANT HEALTH INSPECTION SERVICE					Instructions: Type information requested. Block 1 – assign a number for each collection using your own numbering convention or use the following example by beginning with the year, followed by the collector's initials and the collector's number. Example: 14-JD-001.					LOT NUMBER		PRIORITY <input type="checkbox"/> URGENT <input type="checkbox"/> PROMPT <input type="checkbox"/> ROUTINE	
SPECIMENS FOR DETERMINATION													
1. COLLECTION NUMBER			2A. DATE - SUBMISSION			2B. DATE - COLLECTION			3. SUBMITTING AGENCY			Pest Data Section – Complete Blocks 14, 15 and 16. Complete items 17 and 18 if a trap was used.	
			MONTH DAY YEAR			MONTH DAY YEAR			<input type="checkbox"/> State Cooperator <input type="checkbox"/> University <input type="checkbox"/> APHIS PPO			<input type="checkbox"/> Other:	
4A. NAME OF SUBMITTER				4B. NAME OF COLLECTOR				6. TYPE OF PROPERTY (FARM, RESIDENCE, NURSERY, ETC.)				INTERCEPTION SITE	
5. ADDRESS OF SUBMITTER						7. NAME AND ADDRESS OF PROPERTY OWNER							
ZIP						CITY		COUNTY		STATE			
EMAIL ADDRESS OF SUBMITTER						LATITUDE			LONGITUDE				
8. REASON FOR IDENTIFICATION ("X" all applicable items)													
A. Biological Control (Target Pest Name _____)						E. Export Certification							
B. Damaging Crops/Plants						F. Targeted Survey (Pest Name _____)							
C. Suspected Pest of Regulatory Concern (Explain in REMARKS)						G. Smuggling Interdiction/Trade Compliance (SITC)							
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						11. QUANTITY OF HOST							
NAME OF HOST (Scientific name and name of cultivar if appropriate)						NUMBER OF ACRES/PLANTS			Plant affected (insert figure and indicate)				
						<input type="checkbox"/> Number:			<input type="checkbox"/> Percent:				
12. PLANT DISTRIBUTION				13. PLANT PARTS AFFECTED									
<input type="checkbox"/> Limited				<input type="checkbox"/> Leaves, Upper Surface		<input type="checkbox"/> Trunk/Bark		<input type="checkbox"/> Bulbs, Tubers, Corms		<input type="checkbox"/> Seeds			
<input type="checkbox"/> Scattered				<input type="checkbox"/> Leaves, Lower Surface		<input type="checkbox"/> Branches		<input type="checkbox"/> Buds					
<input type="checkbox"/> Widespread				<input type="checkbox"/> Petiole		<input type="checkbox"/> Growing Tips		<input type="checkbox"/> Flowers					
				<input type="checkbox"/> Stem		<input type="checkbox"/> Roots		<input type="checkbox"/> Fruits or Nuts					
14. PEST DISTRIBUTION				15. <input type="checkbox"/> INSECTS <input type="checkbox"/> NEMATODES <input type="checkbox"/> MOLLUSKS									
<input type="checkbox"/> FEW				NUMBER SUBMITTED		LARVAE	PUPAE	ADULTS	CAST SKINS	EGGS	NYMPHS	JUVS.	CYSTS
<input type="checkbox"/> COMMON				ALIVE									
<input type="checkbox"/> ABUNDANT				DEAD									
<input type="checkbox"/> EXTREME													
16. SAMPLING METHOD				17. TYPE OF TRAP AND LURE				18. TRAP NUMBER					
19. REMARKS										METHOD			
										<input type="checkbox"/> MORPHOLOGY			
										<input type="checkbox"/> SYMPTOM			
										<input type="checkbox"/> CULTURE			
										<input type="checkbox"/> SEROLOGICAL			
										<input type="checkbox"/> PCR			
										<input type="checkbox"/> SEQUENCING			
20. TENTATIVE DETERMINATION						DETERMINED BY			POSITION AND AFFILIATION				
21. FINAL DETERMINATION AND NOTES (Not for Field Use)													
PRINT NAME (Person Making Final Determination)										METHOD			
DISPOSITION OF SPECIMEN/SAMPLE										<input type="checkbox"/> MORPHOLOGY			
<input type="checkbox"/> Returned <input type="checkbox"/> Retained for Collection/Stored <input type="checkbox"/> Destroyed <input type="checkbox"/> Transferred to:										<input type="checkbox"/> SYMPTOM			
SIGNATURE										<input type="checkbox"/> CULTURE			
DATE				LAB CONFORMATION NUMBER				<input type="checkbox"/> SEROLOGICAL		<input type="checkbox"/> PCR			
								<input type="checkbox"/> SEQUENCING		DATE RECEIVED			

PPQ Form 391
AUG 2014

Previous editions are obsolete.

INSTRUCTIONS

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

BLOCK	INSTRUCTIONS
1	<p>1. Assign a number for each collection using your own numbering convention or use the following example by beginning with the year, followed by the collector's initials and the collector's number.</p> <p style="text-align: center;">EXAMPLE In 2014, Brian K Long collected his first specimen of the year for determination. His first collection number is 14-BLK-001</p> <p>2. Enter the collection number</p>
2A-2B	Enter dates
3	Check block to indicate Agency submitting specimens for identification
4A	Enter name of submitter
4B	Enter name of collector
5	Enter address of submitter
6	Enter type of property specimen obtained from (farm, nursery, residence, etc.)
7	Enter name and address of property owner
8A-8H	Check all appropriate blocks
9	Leave Blank
10	Enter scientific name of host, if possible
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	<p>0 Check appropriate block to indicate type of specimen</p> <p>0 Enter number specimens submitted under appropriate column</p>
16	Enter sampling method
17	Enter type of trap and lure
18	Enter trap number
19	Provide a brief explanation if Prompt or URGENT identification is requested
20	Enter a tentative determination and who made it
21	Leave blank

Distribution of PPQ Form 391

Distribute PPQ Form 391 as follows:

1. Send Original along with the sample to your Area Identifier or for national confirmation.
2. Retain and file a copy for your records.