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New Pest Response Guidelines

Ralstonia solanacearum race 3 biovar 2

Southern Wilt of Geranium

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The original version 3 of the Action Plan (New Pest Response Guidelines) for *Ralstonia solanacearum* race 3 biovar 2 was issued February 27, 2003. Previous versions are not to be used in program implementation.

These New Pest Response Guidelines were revised and prepared by:
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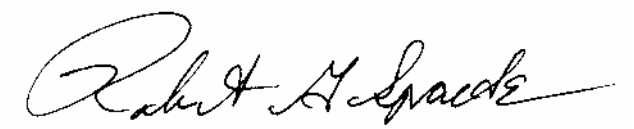

January 14, 2004

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Cover Photo: **Wilting symptoms on geraniums caused by, *Ralstonia solancearum* race 3 biovar 2. Courtesy of the Wisconsin Department of Agriculture, Trade and Consumer Services**

CONTENTS

INTRODUCTION

Purpose

This New Pest Response Guidelines document presents available information for implementing detection, control, containment, or eradication of *Ralstonia solanacearum* race 3 biovar 2. It provides guidelines for designing a program to detect and respond to an infestation on nursery geraniums, other nursery hosts, and solanaceous field crop hosts. Specific emergency program activity should be based on information available at that time.

The United States Department of Agriculture (USDA), Animal and Plant Health Inspection Service (APHIS), Plant Protection and Quarantine (PPQ) agency developed these guidelines through discussion, consultation, or agreement with other APHIS staff, the Agricultural Research Service (ARS), university advisors, States, and industry. It is to be used in conjunction with other agency regulations, guidelines, and manuals when conducting program activities. The information contained in these guidelines is based on the best scientific information available at the time of writing in consultation with States and industry. The guidelines will be updated as new information becomes available. Specific emergency program actions should be based on the best information available at the time of the incident.

Disclaimers

Document Comprehensiveness: This document is not intended to be complete and exhaustive, but provides a foundation, based upon the literature available, to assist further work. Some key articles were not available at the time of writing, and not all specialists and members of the research community were consulted for their advice.

Commercial Suppliers or Products: Any references to commercial suppliers or products should not be construed as an endorsement of the company or product by the USDA.

Ralstonia Infection Prevention

To minimize the effects of this disease and control actions that may be necessary, it is recommended that growers develop and implement effective sanitation procedures to ensure that the pathogen does not spread within their greenhouse or nursery facilities, associated support buildings, equipment or vehicles.

Additionally, federal and state regulatory officers must conduct inspections and apply control measures to ensure that the disease or pathogen does not spread within or between greenhouses or nurseries, associated support buildings, equipment, vehicles, or fields and does not escape into other production systems**. However, since inspectors could inadvertently spread *Ralstonia solanacearum* race 3 biovar 2 or other pathogens through the inspection process, federal and state**

regulatory officers conducting inspections, before entering and upon leaving each greenhouse and nursery location, follow the sanitation guidelines in the beginning of the Survey section to prevent spreading contaminated plant material, soil and/or water to other facilities.

Suggested Practices for Nursery Growers To Lessen the Impact of the Disease and a Quarantine

Practices by growers that can minimize the impact of the disease and of a possible quarantine include:

- Not using sub-irrigation, or shared watering systems;
- Us strict sanitary practices when handling or propagating plants;
- Not placing plants beneath the drip line of hanging geraniums;
- Maintaining varieties of geraniums from foreign sources separate;
- Disinfecting water systems and regularly treating irrigation water from any source;
- Keeping greenhouses, areas around greenhouses, and irrigation water holding or overflow ponds free of weeds.

Growers can be directed to further consult the "Minimum Sanitation Protocols for Offshore Geranium Cutting Production" developed by APHIS and used by off-shore geranium suppliers.

These are available at on the APHIS Ralstonia Pest Alert website at: http://www.aphis.usda.gov/ppq/ep/ralstonia/index.html

Some of the practices in this document may also be useful to domestic growers to help determine what methods might help minimize contamination in specific establishments that grow rooted cuttings in preparation for sale in the United States.

Program Safety

The safety of the public as well as the program personnel is a priority consideration in preprogram planning and training, and throughout program operations. Safety officers and supervisors must enforce on-the-job safety procedures.

Support for Program Decision-Making

The USDA/APHIS/PPQ Center for Plant Health, Science and Technology (CPHST) provides technical support to emergency pest response program directors concerning risk assessments, survey methods, control strategies, and other aspects of pest response programs.

PEST INFORMATION

Systematic Placement

Domain:	Bacteria
Phylum:	Proteobacteria
Class:	Betaproteobacteria
Order:	Burkholderiales
Family:	Ralstoniaceae

Full Name: *Ralstonia solanacearum* (Smith 1896) Yabuuchi et al. 1995 [race 3, biovar 2]

Synonym: *Bacillus solanacearum* (Smith 1896)
Pseudomonas solanacearum (Smith) Smith 1914
Burkholderia solanacearum (Yabuuchi et al. 1992)

Approved common names (APS, 2003):
Bacterial wilt (pepper, potato, tomato)
Southern wilt (geranium)

Additional common names: Bacterial wilt of potato, bacterial wilt of solanaceous crops, brown rot of potato, brown rot of solanaceous crops, southern bacterial blight of tomato, southern bacterial wilt (CABI, 2003).

Background Information

Ralstonia solanacearum is a plant pathogenic bacterium that causes wilt diseases. Various races of this organism affect different crops around the world including tomato, potato, eggplant, banana, and tobacco. Of particular concern to the US is *Ralstonia solanacearum* race 3 biovar 2 because of its affect on eggplant, geraniums, potatoes, and tomatoes. While race 1 is endemic to the Southeastern US where it can affect tomato crops, *Ralstonia solanacearum* race 3 biovar 2 is not known to occur in the US and is considered of quarantine importance.

The bacteria of all races of *Ralstonia solacearum* can be transmitted through contaminated soil, irrigation water, equipment, or personnel. For example, it may be spread by transplanting and propagating infected plants, taking cuttings without disinfecting cutting implements between plants, pinching buds of plants without sanitizing, and especially by shared water irrigation systems. This bacterium can be spread in contaminated soil and on soiled shoes from contaminated areas. Infection occurs typically through the roots and wounding in root areas, a normal physiological process as rootlets grow. Bacteria are normally concentrated in the lower stem portions of the plant. The pathogen does not readily spread from plant-to-plant through the splashing of water, leaf-to leaf contact, or aerially. Spread can be controlled in greenhouses by the application of strict, sound sanitation practices.

Strains of *Ralstonia solanacearum* cause economic damage to diverse agricultural crops and are widespread (Hayward, 1991; Sequeira, 1994). The species is differentiated into five races according to host range (Buddenhagen, Sequeira, and Kellman, 1962), and into biovar classifications according to physiological tests (Hayward, 1964; Hawyard, 1991). Race 1 strains (biovars 1 and 3) have been found to occur on greenhouse ornamentals in the northern hemisphere (CABI, 2003). Race 1 also has a wider host range and is endemic to the southeastern United States.

Table 1 from Daughtrey, 2003 summarizes the host ranges and geographic distribution of the various races, reprinted, with slight modification, from Denny and Hayward, 2001.

Table 1. Races and Biovars of *Ralstonia solanacearum*

Race	Host Range	Geog. Distribution	Biovar
1	Wide	Asia, Australia, Americas	3, 4 1
2	Banana Other Musa spp.	Caribbean, Brazil Phillipines	1
3	Potato, plus other Solanaceae, Geranium; plus others	Worldwide except US and Canada	2
4	Ginger	Asia	3, 4
5	Mulberry	China	5

Race 3 is found worldwide except in the US and Canada. Hosts of *Ralstonia solanacearum* race 3, biovar 2, are usually restricted to cultivated Solanaceous species such as potato and tomato, and has been reported occasionally on *Solanum melongena* (eggplant), *Capsicum annum* and some solanaceous weeds (Martin & French, 1995). A number of additional symptomless weed hosts have also been reported. These weed hosts may enable race 3, biovar 2, to survive in a latent form within the host or in their root areas in the soil (Janse, *et al.*, 2003).

Historical Information

There are some questionable reports of findings of *R. solanacearum* race 3, biovar 2, in some *Pelargonium* spp. from Western Australia (Pittman, 1933) and Tanganyika (Wallace, 1934). In the United States a 1979 study of host tests with a strain of *R. solanacearum* isolated from *Pelargonium x hortorum* (=*P. zonale* hybrids or geranium) indicated the presence of race 3, because the isolate was not pathogenic on tobacco (Strider *et al*. 1981).

In 1999, *Ralstonia solanacearum* race 3, biovar 2, was reported in the US in commercially grown geranium, *Pelargonium zonale* (imported from Guatemala) in the States of New Jersey, New York, Ohio,

Pennsylvania, South Dakota and Wisconsin (Hudelson *et al*. 1999 and 2002; Nameth, 1999; SPRO, 2002). These findings resulted in a review of foreign site production facility practices that export geranium cuttings to the US.

Also in 1999, the bacterium was detected in the United Kingdom on imported geranium cuttings produced in Kenya for the European market (Janse, *et. al*., 2003). During September - December 2000, symptoms of bacterial wilt were observed in several geranium nurseries in Belgium and Germany (Janse, *et al*., 2003).

Recently Kim and Olson (2003) reported a strain of *Ralstonia solanacearum* that they received from Connecticut in 1995, was originally isolated from a geranium plant of Guatemalan origin. They also confirmed the presence of a similar strain on wilted geranium plants from greenhouses in Pennsylvania in 1999 and 2000 and from Delaware in1999, these plants were also imported from Guatemala.

In February of 2003, geranium plants imported from Kenya were implicated as the source of *R. solanacearum* race 3 biovar 2 that resulted in eventual detections in 127 individual nurseries in 27 US states. Imports from Kenya were halted and phytosanitary requirements implemented for all geranium imports from countries that have *R. solanacearum* race 3 biovar 2.

US detections in early 2003 have been within greenhouses and infections were contained, plants destroyed, and the disease eradicated through industry practices or control programs.

In late 2003, a certification program for geraniums produced outside the US was implemented to require specific clean culture practices and regular testing at facilities exporting to the US. Geranium plants exported to the US require phytosanitary certification and a statement that testing showed geraniums shipped to the US to be free of *R. solanacearum* race 3, biovar 2.

Economic Impact

R. solanacearum race 3 biovar 2, is a serious pathogen that causes brown rot, or bacterial wilt, of potatoes (Hayward, 1991; Janse, 1996). Bacterial wilt of potato has been estimated to affect about 3.75 million acres in around 80 countries with global damage estimates currently exceeding $950 million, per annum (DEFRA, 2003). It is adapted to cooler temperatures and would be particularly damaging to potato production regions of the US. While race 1 causes losses to tomato crops in Florida (Momol, et al. 2003), the affect of race 3 on tomatoes and other solanceous crops in the US is unknown.

Hosts

Other than geranium *(Pelargonium* spp.) most other hosts of *R. solanacearum* race 3 biovar 2 are in the family Solanaceae. The major crop host plant species include potato, tomato, peppers and eggplant. Other crop hosts are implicated by laboratory testing. Weed hosts include many nightshades including *Solanum dulcamura* and *S. nigrum*. A complete review published hosts appears in Janse, *et al*., 2003.

Geographic Distribution

Ralstonia solanacearum race 3 causes brown rot of potatoes and also infects tomato and other solanaceous and nonsolanaceous hosts. *R. solanacearum* biovar 2, is commonly referred to as the potato pathogen or the race 3 bacterium. Geographical distribution records listed below are of confirmed race 3 or of presumed race 3 based on pathogenicity to a list of host differentials, which include potato, tomato, and tobacco (CABI, 2003).

Worldwide:

North America: United States and Canada- not known to occur. Present in Mexico (CABI, 2003).

EPPO Region: Belgium, France, Germany, Hungary, Netherlands, Spain, Canary Islands, United Kingdom and Lebanon (CABI, 2003)

Asia: Bangladesh, China (Fujian, Guangdong Guangxi, Hebei, Jiangsu, Taiwan, Zhejiang), India (Himachal Pradesh, Madhya Pradesh, Maharashtra, Manipur, Meghalaya, Tamil Nadu, Tripura, Uttar Pradesh, West Bengal), Indonesia (Java), Iran, Japan (Kyushu), Nepal, Pakistan, Philippines, Sri Lanka (CABI, 2003), Republic of Korea (NIAST, 2002).

Africa: Burundi, Egypt, Kenya, Libya, Réunion, South Africa, Zambia (CABI, 2003)

South America: Argentina, Bolivia, Brazil (Goias, Parana, Pernambuco, Rio Grande do Sul, Santa Catarina, Sao Paulo), Chile, Colombia, Peru, Uruguay (CABI, 2003)

Central America and Caribbean: Costa Rica, Guadeloupe (CABI, 2003)

Oceania: Australia (New South Wales, South Australia, Victoria) Papua New Guinea (CABI, 2003).

SURVEY PROCEDURES

Nursery Hosts: This section contains procedures related to surveying nurseries for infection in geranium and other cultivated hosts.

Sanitation Precautions for Inspectors

When visiting destination nurseries to conduct surveys or to take samples, regulatory officials are to take strict measures to prevent contamination by plant pathogens between greenhouse and nurseries during inspections.

Acceptable methods of protection include the use of disposable gloves and booties changed between visits or an effective antimicrobial soap, lotion, or disinfectant used according to the label instructions.

Wash hands with an approved microbial soap. If not using a microbial soap, wash hands with regular soap and warm water to remove soil and debris. Then use an alcohol-based antimicrobial lotion, such as Purell® or an equivalent product (63% ethyl alcohol). This product has demonstrated efficacy against various resilient *Pseudomonas* species and so is effective against *R. solancearum*. If hands are free of soil or dirt, the lotion can be applied without washing. Unlike some antimicrobial soaps, an antimicrobial lotion is less likely to irritate the hands and thereby improve compliance with hand hygiene recommendations.

If soaps are not used, then disposable latex or nitrile gloves can be used as an alternative, and must be changed between greenhouse visits. Footware should be disinfected using a footbath or other appropriate method when moving between greenhouses or nurseries. If footbaths are not available use disposable booties available from hospital suppliers. Booties must be changed between visits to greenhouses.

General Detection Survey

The purpose of a general detection survey is to determine if a pest exists in an area. This can be extremely broad in scope, as when assessing the presence of the disease in a wide area or it may be restricted to discovering if a specific pest is present in a certain area. Based strictly a negative result in a detection survey, it is not valid to claim that a pest *does not* exist in an area if results are negative. Negative results are valuable, however, for providing clues as to mode of dispersal, temporal occurrence, or industry practices are also important particularly when considered with results from similar areas or proximities.

General detection surveys for geraniums infected by *R. solanacearum* race 3 biovar or other cultivated hosts in nurseries should be conducted by state inspectors in conjunction with federal PPQ inspectors in conducting nursery inspections. PPQ inspectors may inspect nurseries

without a state inspector if they have permission of the nursery owner/manager and have advised the state of their visit.

Symptomology for Inspections During Detection Surveys

Surveys in nursery facilities are conducted visually by looking for plants with typical wilting symptoms of bacterial diseases. The absence of wilt symptoms, however, does not necessarily mean *R. solanacearum* race 3 biovar 3 is not present in the facility. Recent information suggests that some infected plants may not express symptoms, a condition known as latency, even when held for prolonged temperatures.

Wilting symptoms in geraniums caused by *Ralstonia* species are similar to wilting symptoms caused by other bacterial pathogens such as *Xanthomonas campestris* pv. *pelargonii*, the agent of bacterial blight. The primary geranium symptom of infection by *R. solanacearum* race 3 biovar 2, is wilting of leaves and/or abnormal yellowing of lower leaves, while *Xanthomonas campestris* pv. *pelargonii* can also produce leaf spots. Bacterial streaming may be seen if stem sections from *Ralstonia* infected, symptomatic plants are placed into water. If infected with *Ralstonia*, vascular discoloration of the stem is common, and roots may sometimes turn brown. However, with *Xanthomonas campestris* pv. *pelargonii*, vascular discoloration is less pronounced or absent, and roots remain white.

Therefore, when wilting symptoms are present, do not make a field diagnosis such as for *Xanthmonas campestris* pv. *pelargonii*. Always collect a sample for testing at the appropriate diagnostic laboratory. See Appendix 7 for photos of typical wilting symptoms in geraniums caused by both organisms.

Also, visual surveys of other hosts in greenhouses should be conducted during geranium inspections. Symptomology in other infected hosts such as tomato, eggplant, and pepper may also include severe wilting. Some of these host's wilting symptoms are found in Appendix 7. Weed hosts, however, generally do not display wilt symptoms.

In addition, inspect nursery waste areas for discarded plants. Ask the nursery owner/manager to identify cull piles. Examine them for the presence of recently discarded, wilting or dead geranium plants. Discarded host plants that are not completely dried up can still be prepared for sample submission. Document where the sample was taken.

Detection Survey after Initial US Detection

Detection surveys at nurseries after a US detection consists of inspections of suspect geraniums shipments on hold and any other hosts in nurseries suspected of having material at risk for *R. solanacearum* race 3 biovar 2. Inspections are conducted by State, and/or PPQ in

concurrence with State inspectors. Suspect geranium shipments are those held because of their association with a positive detection from another source country, rooting station, or nursery. Be sure to follow sanitation procedures during nursery visits.

Detection survey inspections are to be conducted visually by examining host plants for wilting symptoms, taking samples of symptomatic plants, and having them submitting for testing (see sample collection procedures in the Identification Section).

Nursery owners/managers in the absence of inspection officials that note the presence of plants with wilting symptoms should immediately remove the plants from the bench or rack, mark the location where plants were taken and notify a State or PPQ inspector. The whole plant should be bagged and sealed in double zip-lock bags and then labeled with the date, name of person responsible, and location where plant was taken. These plants need to be kept under refrigeration or in cool conditions until the inspector arrives (instruct managers not to freeze the sample).

Random sampling of geranium plants not showing symptoms is not recommended at this time because of a lack of sampling methodology and processing capability.

Monitoring Surveys

Once a nursery with positive confirmed infection of *R. solanacearum* race 3 biovar 2 has all suspect geraniums and potentially infected plants destroyed and affected areas disinfected, some kind of follow-up monitoring survey is necessary. Be sure to follow sanitation procedures during nursery visits in a monitoring survey.

Inspectional visits during the same, or subsequent growing seasons is appropriate in order to examine hosts for symptoms. If sampling protocols are available, water sampling of irrigation systems or ponds may also be included with additional inspections of host crop areas in runoff areas adjacent to positive testing nurseries.

<u>Solanaceous Crops:</u> This section contains guidelines for surveying host crops such as potato, tomatoes, eggplant, and pepper for infection.

In Development

DIAGNOSTICS AND IDENTIFICATION

Importance

Accurate identification of the quarantine pest is pivotal to assessing its potential risk, developing a survey strategy, and deciding the level and manner of control.

Authorities

For this organism, State and cooperating university diagnostic laboratories can make determinations of bacterial infections to the level of genus and species, i.e., *Ralstonia solanacearum*.

All laboratories knowingly possessing cultures of *R. solanacearum* race 3 biovar 2 must follow procedures listed in the Federal regulation on *Agricultural Bioterrorism Protection Act of 2002* (7 CFR Part 331). Failure to comply can result in severe penalties. (See Permitting information at then end of this section and consult the PPQ permits website for further information on diagnostic laboratory responsibilities regarding the handling of select agents).

A USDA-recognized national authority for the quarantine significant taxon must positively identify the suspected pest before initiation of any program quarantine activities. In the case of *R. solanacearum* race 3 biovar 2, the final identification to this race and biovar is performed at the National Plant Germplasm and Biotechnology Laboratory in Beltsville, Maryland (CPHST Beltsville Lab). This laboratory also has all the necessary registrations and containment approvals to handle select agents.

Geranium Symptom Identification

Some pre-identification and screening can be performed by field personnel assigned to the program. Only plants exhibiting wilting symptoms are to be sampled and submitted. Consult the survey section for more information on symptomology and see examples in Appendix 7. Other bacterial diseases can also cause wilting symptoms, so screening by diagnostic laboratories is a necessary and required step. These must be State, cooperating university, National Plant Diagnosis Network laboratories, or recognized permitted private laboratories listed in Appendix 4.

Nursery Hosts Sample Collection Procedures

For sampling of plants showing wilting symptoms, the whole plant must be collected including roots, preferably without soil. Bare root plants are ideal. **Since the pathogen is concentrated in the lower stem, the disease may not be detected from samples if only leaf or partial stem samples are taken.**

Place the plant sample in a zip lock bag and seal it. On this bag holding the sample, write the sample number with a Sharpie®

permanent pen. Place this bag inside a second zip-lock sample bag with a completed Sample Accompaniment Form (see Appendix 2). Legibly fill in the information on the form as completely as possible.

If plants in soil are submitted, assure that you place a separate plastic bag around the pot with a rubber band around plant base to restrict soil spillage and contamination of the plant tissue. If soil is left over, dispose of it properly.

Entire samples of plants should be submitted, not sub-samples. Samples must include lower stems at a minimum and samples should not consist of detached leaves and/or detached petioles. Samples that are necrotic (brown) or fermented upon arrival cannot be tested and will be rejected.

Sample Labeling, Numbering, and Record Keeping

The inside bag and Sample Accompaniment form must have sample numbers generated in the field recorded on them. Assign and record for each sample a unique ID number of the following format:

XX-ABC-0001

where XX is your two letter state code, ABC is a three letter, state assigned facility code, and 0001 is the sample number for that facility. It is very important that unique sample numbers are used for each sample. Keep a log of assigned sample numbers.

If diagnostic screening laboratories are part of the National Plant Diagnosis Network (NPDN), samples may be submitted using an NPDN number format. If a bar-coded sample number system is available, be sure to include the bar-code in the sample on the PPQ form 391.

Complete a muticopy carbon PPQ form 391 marked "URGENT" (use form in Attachment 2 if carboned form not available) with the sample number clearly written. Include a copy with each sample.

Inspectors must provide the sample date with all relevant collection information to with their State Plant Regulatory Official and/or State Plant Health Director as soon as possible. This information should be communicated within a State and with the regional office program contact. If a *Ralstonia* sample tracking database is available at the time of the detection, please enter collection information in the system as soon as possible.

Sample Storage and Forwarding

Samples must be forwarded to approved diagnostic screening laboratories (see Appendix 4) as soon as possible to prevent deterioration of plant tissue.

If storage is necessary, samples in double zip-lock bags as described above are placed in a cooled location or container until ready for shipping. Do not place samples in a freezer. Samples may be held in a standard refrigerator at 34°F (4°C) or at room temperature if less than or 60°F (15°C).

Samples must be sent by overnight delivery. Ice packs are not needed or recommended. Packaging of all samples should be in a larger bag and made leak proof, then must be placed in a sturdy cardboard outer box with insulation to prevent movement within the box during shipping.

Diagnostic Screening Laboratories

To determine if the infection is due to *R. solanacearum,* samples of plants with symptoms taken by field personnel must be first submitted through their normal regulatory networks, i.e., the State's or cooperating university diagnostic laboratory (see a listing in Appendix 4). These labs will perform diagnostics to the genus and species level. They will not determine to the race and biovar.

Diagnostic screening laboratories receiving samples are to communicate the date of receipt with their State Plant Regulatory Official and/or State Plant Health Director. All relevant sample information and the diagnostic lab's species determination must be communicated as soon as possible within a State and with the PPQ regional office program contact.

If a *Ralstonia* sample tracking system is used, at a minimum enter the date received in the system, test results, the determination, date identification made, and who made the identification.

Required Permits

For inspectors shipping plant pest sample material to other labs to test for the presence *R. solanacearum* (*i.e*., identification to species only, not biovar and race), no permit is required for **intrastate** (within the state) shipment. Persons shipping plant material to a diagnostic laboratory in their own state do not need to obtain a PPQ Permit under either the *Plant Protection Act* (Regulations at 7 CFR Part 330, the Plant Pest Permit (PPQ Form 526).

And although **intrastate** transfers of known *R. solanacearum* race 3 biovar 2 are regulated under the *Agricultural Bioterrorism Protection Act of 2002* (Regulations at 7 CFR Part 331), the submission of unknown samples or those only identified to *R.*

solanacerum (not to race and biovar), are not regulated under this Act.

States that do not have their own State department of agriculture or cooperating university laboratory for screening to genus and species may ship **interstate** (across state lines) to use another state's laboratory.

For **interstate** shipping, the originating State must assure, however, that the laboratory in the destination state has the necessary plant pest permit (PPQ Plant Pest form 526) to receive interstate samples. For this purpose, the *Agricultural Bioterrorism Protection Act of 2002* registrations are not necessary. As with intrastate submitted samples to diagnostic labs, interstate shipment of unknown potentially infected plants is not subject to regulations under this Act.

Any laboratory that diagnosis samples to *R. solanacearum* race 3 biovar 2, is required under the *Agriculture Bioterrorism Protection Act of 2002* to immiately notify the PPQ Biological and Technical Services (BTS) Permit Unit regardless of whether a Plant Pest Permit is required. The laboratory possessing the sample has seven days in which to destroy the sample, must give PPQ the opportunity to witness the destruction, and laboratory director complete a form APHIS form 240.

For further guidance on permitting of plant pest material, consult the PPQ permit website at: http://www.aphis.usda.gov/ppq/permits/ or contact PPQ BTS Permit Services on (301) 734-7211, 6828 or 5055.

Alternative Diagnostic Laboratory

The National Plant Diagnosis Network labs (see Appendix 4) can make screen to genus and species for states that prefer not to or cannot screen their own samples. Any sample testing positive for *R. solanacearum* will be forwarded to the USDA, APHIS, PPQ-CPHST lab in Beltsville, MD for confirmatory testing

As another alternative for states that cannot screen their own samples or have not been able to make arrangements with another permitted laboratory out of state, there is currently one private laboratory that already has the necessary permits and authorizations to screen to species, (not race 3 biovar 2). Any sample testing positive for *R. solanacearum* will be forwarded to the USDA, APHIS, PPQ-CPHST lab in Beltsville, MD for confirmatory testing.

That laboratory is: Agdia Inc.,
30380 County Road 6,
Elkhart, IN 46514
phone number: (574) 264-2014, or 1-800-62-AGDIA
www.agdia.com
Other private laboratories may be eligible as permits are approved and these establishment names will be provided.

Diagnostic Tests to Determine Genus and Species

There are a variety of serological tests available to screen for this organism to the genus and species level at diagnostic laboratories. No special permits are needed by these labs to do this test unless they are receiving samples from another state(s).

The three serological tests validated and recognized by the National Plant Germplasm and Biotechnology Laboratory are listed below. See Appendix 3 for more information on these kits.

Validated Screening Test Kit Availability:

The USDA-APHIS-PPQ-CPHST Laboratory in Beltsville, MD, evaluated three rapid serological tests for detection of *Ralstonia solanacearum.* None of the serological tests evaluated will determine the organism to race/biovar.

The three tests evaluated are:

***Rs* ImmunoStrip Test**
Agdia, Inc
30380 County Road 6
Elkhart, IN 46514
www.agdia.com
Ph. 800-622-4342
FX 219-264-2153

Potato Brown Rot Pocket™ Diagnostic
Central Science Laboratory (CSL)
Sand Hutton, York, YO41 1LZ
www.csl.gov.uk
Ph 44 1904 462600
FX 44 1904 46211

***Ralstonia solanacearum* SPOT√CHECK LF™**
ADGEN, LTD.
Nellie's Gate, AYR
Scotland, KA6 5AW
www.adgen.co.uk
Ph 44 1292 525275
Fx 44 1292 5255477

ELISA Test Kits

There are also ELISA (Enzyme Linked Immunosorbant Assay) kits tested and validated by the CPHST Beltsville lab that will also make diagnoses to genus and species. These are listed in Appendix 3 with strengths and sensitivities of each listed.

Culturing

Diagnostic laboratories can forward cultures after screening to genus and species has been performed. See Appendix 3 for recommended methods of culturing. When sending cultures for race and biovar determination, the sender must assure that packaging is secure as with plant samples.

Approved Laboratory for Confirmatory Testing to Race and Biovar

Once the plant material has been screened and is known to contain *R. solanacearum,* **forward the sample or culture as soon as possible by overnight carrier to the CPHST Beltsville laboratory for confirmation to race and biovar**.

There is only one diagnostic laboratory with the proper permits to test for the presence of *Ralstonia solanacearum* race 3, biovar 2. This is the USDA, APHIS, PPQ-CPHST Beltsville laboratory (*address below*) whose Director, Dr. Laurene Levy, has all necessary authorizations to receive samples submitted for diagnostics to the race and biovar level.

Attention: DeVries/Levy
USDA, APHIS, PPQ-CPHST
BARC-East, Bldg. 580
Powder Mill Road
Beltsville, MD 20705
phone number: 301-504-7100
fax number: 301-504-8539

The APHIS PPQ CPHST lab also wants the name of the kit used for the detection and specific descriptive information on the test results (i.e., intensity of the test band, OD of the ELISA well, or digital images of the test result).

Sample Condition for Submitting for Confirmatory Testing

The following information regarding samples should be noted when forwarding *R. solanacearum* suspect samples to the APHIS, CPHST lab in Beltsville, MD:

Entire samples of suspect *R. solanacearum* positives should be submitted to the Beltsville lab, not sub-samples. Samples need to include lower stems. Samples should not consist of detached leaves and/or detached petioles. If samples are necrotic (brown) or fermented upon arrival in Beltsville, they will be rejected.

Completing the PPQ form 391 Determination Section

Diagnostic screening laboratories must write their determinations for each sample on the PPQ form 391 with the name and phone number of the responsible diagnostician, keep a copy, and follow the same sample packaging instructions as above.

The APHIS Beltsville staff requests the following additional information please be noted on the 391 form:

a) The test kit used to make the *R. solanacearum* diagnosis.
b) Information whether the *R. solanacearum* test was strongly positive or weakly positive, etc. If possible, a print-out of a digital image of the stick or plate should accompany the sample and paperwork.
c) What specific plant part tested positive? (symptomatic and/or lower stem area should be the only tissue test

Please call or fax to the above laboratory numbers to notify the CPHST Beltsville laboratory that you are sending samples of *Ralstonia solanacearum.* (If faxing, include all pertinent information and contact name and number).

Sample Packaging and Documentation

Diagnostic screening laboratories must send plant samples or cultures by overnight delivery. Ice packs are not needed or recommended. Packaging of all samples should be in a larger bag and made leak proof, then must be placed in a sturdy cardboard outer box with insulation to prevent movement within the box during shipping.

Include the completed PPQ form 391, a copy of the Sample Accompaniment Form, and copies of any relevant tags or barcodes that came with the sample.

Saturday Delivery

Only with specific permission from APHIS PPQ headquarters should samples be sent on Fridays by FedEx®. Although it is possible to have Saturday delivery by overnight carriers to the Beltsville facility, often samples are not delivered even with special arrangement with FedEx®. However the laboratory will be operating on Saturday if it becomes necessary, and only will occur if a large program is being conducted requiring additional hours of operation. If you have obtained permission from APHIS PPQ headquarters to send samples for Saturday delivery, please send the FedEx® tracking number to Beltsville via email to Laurene Levy (lauren.e.levy@aphis.usda.gov) and Renee DeVries (renee.m.devries@aphis.usda.gov) so they can notify their Fedex® local office to authorize Saturday delivery**.**

Notification of State Officials of Sample Submissions and Results

Notify the State Plant Health Director and State Plant Regulatory Officals in the sample state of origin and fax the PPQ regional office of any sample forwarding information, completed documentation, including overnight freight tracking information. Once results are known, States will be notified by the PPQ regional office of the results.

Please do not call the Beltsville laboratory to get sample results, this information will be reported to the regions and States from headquarters as soon as they are available.

Sample Processing Time

Growers and cooperators need to be aware that sample processing time of 48 hours to three weeks depending on the sample condition and the ease with which diagnostic tests can be performed. This is in addition to the time it takes process and forward samples from the intermediate state and cooperating university diagnostic laboratories.

Special Instructions to Diagnostic Laboratories Having Positive Confirmations

In compliance with the *Agricultural Bioterrorism Protection Act of 2002* (7 CFR Part 331) if a diagnostic laboratory held back part of a screened sample or culture for voucher purposes and that sample the sample forwarded to the USDA Beltsville Laboratory came back as positive for *Ralstonia solanacearum* race 3 biovar 2, the diagnostic laboratory is required to notify PPQ BTS Permit Unit that the sample exists. Destruction must take place within seven (7) days of notification that the sample is positive for *R. solanacearum* race 3 biovar 2 and a PPQ representative must have the opportunity to witness the destruction of the sample or culture within that time period. The responsible laboratory manager/plant pathologist must also complete the APHIS form 2040 available on the Permits website at:
http://www.aphis.usda.gov/ppq/permits/agr_bioterrorism/index.html

The APHIS PPQ CPHST lab in Beltsville, has the necessary registrations to possess this select agent, and is also required by law to report the positive confirmation within 24 hours of detection, and must submit within 7 days an APHIS 2040 to the PPQ permit unit in Riverdale, MD.

REGULATORY PROCEDURES

<u>Nursery Hosts:</u> The sections below refer to procedures for regulating geraniums and other hosts in nursery establishments.

Instructions To Officers

Agricultural officers must follow instructions for regulatory control measures, treatments or other procedures when authorizing the movement of regulated articles. A full understanding of the instructions and procedures is essential when explaining procedures to persons interested in moving articles affected by the quarantine and regulations. Only authorized treatments may be used in accordance with labeling restrictions. During all nursery visits, please assure that proper sanitation procedures are followed as outlined in the Survey section.

Overview of Regulatory Program for Greenhouse Geraniums after a US Detection

Once a greenhouse detections on geraniums are made and positively confirmed as *R. solanacearum* race 3 biovar 2 in the US, it is necessary determine the origin of the infections and to survey the extent of distribution of potentially infected plants. To determine the origin, a traceback investigation is conducted to determine the foreign source of infection and then the extent of distribution of plants in the US from that source facility. Similarly, traceforward investigations will be necessary to determine if potentially infected plant material had been distributed to other retail locations.

Investigations at traceforward US geranium propagation facilities and their customers will be conducted. Results of investigations will allow implemention of a program of holding varieties or shipments that originated at a particular foreign facility during a shipping period. The irrigation methods, handling, and sanitation practices of each nursery will help assess if additional associated plant material is at risk of infection. Information gathered in the investigation portion will be critical to determining what additional plants to hold and subsequent actions taken. Where guidance is needed, regional contacts will be consulted.

There is no treatment for plants infected by *R. solanacearum* race 3 biovar 2. All plants determined to be regulated and the associated soil will be destroyed and areas sanitized before facilities can be released from quarantine.

If new information indicates a more widespread infestation problem than originally assessed, or new information becomes available, these regulatory guidelines are subject to change.

Investigations Conducted in a Detection Survey after Initial US Detection

A list of nursery facilities will be compiled of geranium varieties and/or shipping windows that have been associated with positively tested, foreign origin geraniums. These lists will be distributed by state to the field offices, and are not to be shared with individuals outside our regulatory cooperators. **Company names and locations on these lists are strictly confidential, and any distribution of lists beyond appropriate regulatory agency contacts is prohibited.**

When notifying nurseries on the list, be sure to identify yourself as a USDA or state regulatory official conducting an investigation of nurseries that may have received *R. solanacearum* infected material. Speak to the nursery owner or manager.

It is important to understand that nursery facilities handling foreign geraniums are of several types. Companies ship cuttings and callused plants through PPQ Plant Inspection stations. The cleared material is then either sent to rooting stations or to direct ship customers which are wholesalers. Both types of facilities grow the plants for a period of time (usually 5 weeks) before being distributed to other entities. Rooting stations, under contract with the company, then ship orders to a variety of customer types including wholesalers and both large and small retail customers. Direct ship facilities can have their own network of retail stores or sell plants to other customers. Handling and cultural practices vary at all steps in the distribution.

During the investigation, request that the nursery owner or manager produce any tags, invoices, shipping lists associated with the positive tested suspect plants on the distributed list. Make copies of these records and locate the suspect plants still on the premises. Take photographs of the area where suspect geraniums are located and any greenhouse conditions including the proximity of neighboring plants that may indicate broader contamination. Make a record of the number of plants, type, and location including an accounting of suspect plants from that shipment not on the premises.

Traceback information gathered from plant tags and invoices can be used to determine the origin of the plants. With timely submitted records from plant distribution companies such as rooting stations or wholesale growers, PPQ headquarters Pest Detection and Management Programs staff can generate lists of nurseries with implicated plant varieties. Considerable efforts have been made by the geranium industry to use bar-coding for plants and cuttings tracking that may have use in these investigations.

This information can be used to determine the extent of distribution of suspect geraniums at various locations in the US. Coordinate with your

State contacts and regional office to gather this information. Additional resources may be needed to quickly gather this information and analyze it in order to hold suspect geranium shipments and potentially infected plants.

Further Investigations at Nurseries with Suspect Geraniums

Once a list of nurseries in the state is generated after the initial US detection, it will be necessary to visit the locations to assess the extent of plant distribution and nursery irrigation and sanitation practices. Various greenhouse practices will prevent or facilitate the spread of bacteria of *R. solanacearum* race 3 biovar 2. Information must be gathered to determine what additional potentially infected plants or areas may be at risk for carrying the pathogen. For more information, refer to the "Plants to Hold" section below.

Examine the type of irrigation system and practices used at the nursery:

- Are plants under a sub-irrigation system where water floods the pots and penetrates from below?
- Do you notice hanging geraniums from suspect shipments? Were other plants (hosts or otherwise) under their drip line?
- Are cuttings of plants being made to propagate more plants?
- Do workers exhibit sanitary handling of plant material?

This kind of observational information should be documented by the inspector. In addition, interview the nursery owner/manager and ask if they have seen wilting geraniums or other hosts during the season in question. Ask if any wilting plants have been discarded and if so, where? Photographs of the facility and written observations can be useful in the investigation phase.

Ask the nursery owner/manager to complete and sign the questionnaire in Appendix 1.

The information gathered in the above questions and documentation will assist in determining if additional plants must be held. Consult with your regional contacts when there are questions concerning a particular facility practice that may affect the number of additional potentially infected plants held and the extent to which that practice is used.

Regulatory Program Definitions

The following are definitions of terms used in the regulatory and other sections of these guidelines, and help determine how we deal with various categories of plants.

Suspect geraniums - geraniums (*Pelargonium* spp.) that are associated with positive testing geraniums. These may be a particular implicated variety or shipment that originated from a rooting station, direct ship facility, or another nursery.

Suspect Shipment - all host plants that were prepared for shipment and transported with suspect geraniums from a common supplier (listed on the invoice or packing list).

Destination nursery - Any nursery identified in traceforward investigations as receiving suspect geraniums or suspect plants, from a rooting station or other nursery.

Positive testing geraniums – geraniums in which *Ralstonia solanacearum* race 3 biovar 2 has been confirmed to be present by the USDA-PPQ-CPHST laboratory in Beltsville, MD.

Potentially infected plants – within the nursery property, these include:

a) all plants under the drip-line of hanging suspect geraniums;
b) other plants that have been planted in the same pots with suspect geraniums;
c) all plants on a shared water irrigation system with suspect geraniums; (*Examples include ebb-and-flow, flood, sub-irrigation, or systems that lack of backflow prevention*).
d) all host plants that may have been infected by a positive testing suspect geranium shipment through unsanitary greenhouse practices. (*Examples include failure to disinfect frequently tools, hands, or equipment during grafting, pruning, de-budding, or de-leafing between varieties, plants are sitting on the ground with a non-porous surface such that puddling may occur under plants).*

Placing Holds at Nurseries

After a destination nursery is identified as having received plants from another source associated with positive testing geraniums, it is important to immediately place holds on all geranium varieties named on distributed lists and initially, any other hosts plants in the facility. If the extent of potentially infected plants at that facility can be ascertained at that time, those not determined to be potentially contaminated can be immediately released.

A "hold" is interpreted as the prohibition of movement of those plants from the property until further evaluations can be made and control actions can be taken on suspect geraniums and other potentially infected plants.

After investigations are made, unless general contamination is suspected, the hold is **not** to be interpreted to:

1) prohibit the movement of all plants in a greenhouse;
2) that plants not held cannot be cared for in a normal and sanitary manner;
3) prohibit the relocation of suspect geranium shipments and potentially

infected plants to a segregated area away from other plants. If the nursery owner/manager wishes to move held plants, assess the risk of such movement and keep records of where relocated plants are.

Record Keeping

Record keeping and documentation is important for any holds and subsequent actions taken. Rely on shipping records and information provided by the nursery owner/manager for how many plants remain, how plants have moved within the nursery, destination of plants sold, and cultural practices employed.

Keep a detailed accounting of the numbers and types of each plant variety held and/or destroyed in control actions. Consult a master list of varieties distributed with the lists of facilities. Draw maps of the greenhouse layout to located suspect geraniums, other potentially infected plants, and water runoff areas including recirculating pond locations. Take photographs of the facility layout, geranium placement, watering method, materials and methods used, plant labeling, and any other situations that may be useful for documentation and analysis.

Keep all written records filed with Emergency Action Notification (EAN, PPQ form 523) copies, copies of sample submission forms, documentation of control activities, and related State issued documents if available.

Plants to Hold

Use the EAN from to place holds at destination nurseries for the following classes of plants:

1) **Suspect geraniums**; which may include:

*Variety level**:*** Depending on the particular program directions, a particular variety or varieties of geranium plants may be considered suspect because they tested positive and traceback investigations to a particular foreign facility determined their overall infection risk. The variety level hold can be most appropriate at rooting stations or direct shipped growers that are large operations whose practice is to keep varieties separate.

Shipment level: Depending on program directions and practices at a previous facility in the distribution, a level of suspect geraniums to hold may include the shipment of all host plants received from that facility.

Release all plants that are **not** considered potentially infected. The following are subject to holds:

2) **Potentially infected plants**- ***High Risk***

Results of investigations at nursery facilities and answers to the questionnaire will determine the level of additional holds of potentially infected plants. After investigations are made, some of these conditions

(continued) **Potentially infected plants- *High Risk***

will require individual judgment and consultation with your regional office. The categories that define higher risk for potentially infecting plants include:

a) Plants beneath the drip line of suspect geraniums – because *R. solanacearum* bacteria are easily shed in water, plants, regardless of species, directly below hanging suspect geraniums are at high risk of soil contamination so must be included. This includes hosts and non-hosts. This is also true for plants stored under benches with suspect geraniums.

b) Plants in the same pots with suspect geraniums – Infected geraniums in the same pots with other plants can easily infect them through water, soil, or root contact. This includes hosts and non-hosts.

c) Plant propagation from suspect geraniums has occurred – Any plants propagated from suspect geraniums are at high risk for infection.

d) Plants on a shared irrigation system with suspect geraniums- the most efficient method of spread for *R. solanacearum* is through water that came in contact with infected plants. It is therefore necessary to hold all plants (hosts and non-hosts) that are on irrigation systems that allows for water flow from one plant to the next. These are various irrigation situations to look for:

i) Sub-irrigation, ebb and flow, or flood irrigation: pots sit in a pan and are irrigated by flooding the pan in various ways.

ii) Backflow prevention: nearly all irrigation systems have check valves to as a safeguard to prevent contaminated water from backing up into the general water supply. If systems lack backflow prevention, there is a high risk of general contamination throughout the system from infected material.

e) Plants in facilities where sanitary cultural practices are not in place – Using grafting knives for making cuttings or grooming plants without disinfection between varieties is a high risk factor in transmitting the pathogen to other host plant lots.

f) Plants placed on the ground, on plastic sheeting or other material that allows puddling between plants - Because the pathogen spreads by water, there is a risk that puddling between plants will cause uptake of bacteria by health plants in the vicinity of infected. The extent to which this occurs needs to be assessed by observation and further evaluations or monitoring may be necessary.

3) Potentially infected plants – *Reduced Risk*

The following greenhouse cultural practices are considered not as high of risk for spreading the pathogen to other plants. Evaluations can be made by the inspector as to the degree to which such practices may take place based on observations and responses to the questionnaire**. It is recommended that the inspector consult with the supervisor, SPHD, SPRO or regional office if there is reason to believe the extent of these practices presents a greater risk and therefore requires additional destruction of plants.** In some cases, after consultation, the degree to which these risk factors are practiced at the facility may determine the need for wider destruction of potentially infected plants, however, in most cases, these nurseries with plants in these reduced risk categories may be held and subject to follow-up inspections to inspect for wilt symptoms.

a) Plants that were not segregated from suspect geraniums – recent information shows that leaf-to-leaf contact between geraniums is not a viable method of pathogen spread, so there is little to no risk of plants in the same proximity of infected plants becoming contaminated by contact. An exception to this might be plants immediately adjacent to infected plants that were watered by hand (using a hose or a water-wand) where splash may transfer the pathogen to the soil in adjacent plants. Generally, drip irrigation and mist systems do not create a risk of splash to adjacent plants. When in doubt, consult your SPHD, SPRO, and regional contact.

b) Plants that were pinched, deleafed, disbudded by hand – this method of grooming presents less risk of spread to non-infected hosts plants than by the use of cutting tools, but some degree of sanitation between varieties is necessary. An exception to this is if little to no sanitary practices are in place for workers who perform plant grooming by hand and sanitation of hands should take place at least between varieties.

Issuing an Emergency Action Notification

An EAN is issued to hold all host plants at facilities that have the suspect geranium varieties and potentially infected plants connected to positive confirmations. Once an investigation determines there are plants that are not suspect geraniums or potentially infected, they may be released and documented on the EAN. The EAN may also be issued to hold plants when wilt is discovered pending positive identification. When a decision to destroy plants is made, or in the case of submitted samples, once positive confirmation is received, the same EAN for which the plants are on hold is used to also document any actions taken such as destruction and disinfection. If varieties or shipments are to be held as separate units, it is advisable to issue separate EAN's for each

held unit of suspect geraniums and potentially infected plants associated with that unit. Electronic EAN's can be used but follow directions in Appendix 8. EAN's are issued under the authority given in 7 CFR 330. It is advised that States issue their own hold orders parallel to the EAN to assure plants cannot move intrastate.

Quarantine Actions

If necessary, the Deputy Administrator will issue a letter directing PPQ field offices to initiate specific actions under the Plant Protection Act until emergency regulations can be published in the Federal Register.

The Plant Protection Act of 2000 provides for authority for emergency quarantine action. This provision is for interstate regulatory action only; intrastate regulatory action is provided under State authority. However, if the Secretary of Agriculture determines that an extraordinary emergency exists and that the measures taken by the State are inadequate, USDA can take intrastate regulatory action provided that the Governor of the State has been consulted and a notice has been published in the Federal Register. If intrastate action cannot or will not be taken by a State, the PPQ may find it necessary to quarantine an entire state.

A General Memorandum of Understanding between States and PPQ exists for each State and, in certain circumstances, may facilitate access to private property, in the absence of landowner permission, by PPQ Officers in conjunction with State inspectors to place facilities under notification and witness actions specified in the emergency action notification. Check with your SPHD for clarification.

Quarantine Actions Required

After investigations are performed (and additional consultations made if necessary), the unit of held suspect geraniums and potentially infected plants associated with them must be destroyed /disposed of and the area disinfected under the conditions described in the Control Section.

Regulated Articles

Host Plants: Cultivated Plants, cuttings, or parts of the following hosts of *Ralstonia solanacearum* race 3 biovar 2 are regulated if tested positive and considered potentially infected:

Table 2. Cultivated Hosts of *R. solanacearum* race 3 biovar 2

Common Name	Scientific Name
Geranium	**Pelargonium* spp.
Tomato	*Lycopersicon esculentum*
Peppers	*Capsicum spp.*
Eggplant	*Solanum melongena*
Potato	*Solanum tuberosum*
Bean	*Phaseolus vulgaris*
Bittergourd	*Momoridica charantia*
Beet	*Beta vulgaris*

*consult Appendix 6 for the *Pelargonium* species used most often in trade

Host Plants: Weeds Weed hosts of *R. solanacearum* race 3 biovar 2 are often symptomless but must be destroyed in greenhouses with positive detections including an area within one (1) meter around the outside perimeter of the greenhouse. Follow destruction/disposal and disinfection procedures in the Control Section.

Table 3. Weed Hosts of *R. solanacearum* race 3 biovar 2

Common Name	Scientific Name
Black nightshade	*Solanum nigrum*
Climbing nightshade	*Solanum dulcamara*
Horsenettle	*Solanum carolinense*
Jimson weed	*Datura stramonium*
Purslane	*Portulaca oleracea*
Mustards	*Brassica* spp.
Lambsquarters	*Chenopodium album*
Bittergourd	*Momoridica charantia*

Pots and Media

Pots and soil, planting or rooting media that has been in contact with positive tested plant shipments and other potentially infected plant material are also regulated and subject to destruction, disposal, and disinfection procedures found in the Control Section.

Tools and Equipment

Tools, implements, equipment, benches, greenhouses used in cultivation that may have contacted positive tested plant material are subject to disinfection according to the procedures described in the Control Section.

Other regulated articles include any other products, articles, or means of conveyance, of any character whatsoever, when it is determined by an inspector that they present a hazard of spread of *R. solanacearum* race 3 biovar 2 and the person in possession thereof has been so notified.

Removing Areas from Quarantine

Plants held can be released from Emergency Action Notification conditions if any of the following can be demonstrated:

1. No suspect geraniums from specified suspect rooting and distribution facilities were received at the destination nursery during the specified time periods; **OR**

2. No other host plants from the facility have shown wilt symptoms, or if wilt was detected, they were subsequently sampled, tested, and found negative for *Ralstonia solanacearum* race 3 biovar 2, **and**

3. Suspect geraniums associated with positive confirmed detections of *R. solanacearum* race 3 biovar 2 were disposed of or destroyed along with associated potentially infected plants according to procedures

	described in the Control section. This includes removal and destruction of weed hosts and the disinfection of nursery areas where destroyed plants were held including the tools and other equipment that may have come in contact with suspect geraniums and potentially contaminated plants. Additionally, before release, further traceback or traceforward investigations should not indicate a risk of dissemination of *Ralstonia solanacearum* race 3 biovar 2 to other parts of the nursery property. Before release of a facility, PPQ Officers must conduct a monitoring inspection and document on the EAN that no positives for *Ralstonia solanacearum* race 3 biovar 2 were detected or, if positives were detected, then all destruction, disposal, and disinfection actions taken with dates taken, location, and witnessed by whom. Notify nursery owner/managers that their facilities may be subject to additional monitoring by State or Federal officials for the presence of *Ralstonia solanacearum* race 3 biovar 2.
Regulatory Records	Maintain standardized regulatory records and database(s) in sufficient detail to carry out an effective, efficient, and responsible regulatory program.
Use of Chemicals	The PPQ Treatment Manual and this Guideline identify the authorized chemicals, and describe the methods and rates of application, and any special application instructions. See the Control section for more information. Concurrence by PPQ is necessary before using any other chemical or procedure for regulatory purposes.

<u>Solanaceous Field Crops</u>: **This section describes regulatory options for infections found in potato, tomato, eggplant or pepper crops in outdoor, cultivated situations.**
In development

CONTROL PROCEDURES
Nursery Hosts

Overview

Plant Protection and Quarantine develops and makes control measures available to involved states. Environmental Protection Agency (EPA) approved and labeled treatments will be recommended when available. If additional treatments selected are not labeled for use against the organism or in a particular environment, an emergency exemption can be requested and obtained under Section 18, or 24(c), special local need (SLH), of FIFRA (Federal Insecticide, Fungicide, and Rodenticide Act), as amended. Disinfections conducted with *Ralstonia solanacearum* as a target pest use a FIFRA Section 2ee until that name is specifically added to the label. This designation is necessary because *Pseudomonas* is labeled on recommended products and this is the previous genus name synonymous with *Ralstonia*.

Control Decisions and Oversight

All quarantine actions related to destruction are to be witnessed, supervised, and documented by a PPQ Officer whenever possible. Because *R. solanacearum* race 3 biovar 2 is listed as select agent under the *Agriculture Bioterrorism Protection Act of 2000*, proper supervision and documentation of destructions of infected plant material is critical. If a PPQ Officer is not available, a State cooperating inspector can witness and document the disposal. Disinfections can be witnessed and documented by PPQ Officers or State cooperators.

Labeling

Pesticides, including disinfectants may be applied legally only to those sites specifically listed on the label. It is a violation of Federal law to use a pesticide in a manner inconsistent with the label.

While the proposed chemicals below are approved for an effective eradication program, these chemicals may not be labeled, at the time of a pest detection, for the specific use-site where treatment is required. The prescribed pesticide must be labeled for use on the site where the chemical is to be applied and must be registered for use in the State where the eradication program is occurring. All applicable label directions must be followed, including requirements for personal protection equipment, maximum treatment rates, storage and disposal.

Positive Testing Geranium Lots and Associated Plants

For shipments of suspect geraniums that have tested positive for *R. solanacearum* race 3 biovar 2, place all suspect geraniums and all associated potentially infected plants in double plastic bags and seal for disposal or destruction. This includes the pots, soil or media, and any other item in contact with the plants or soil. Host weeds in positive tested facilities must also be removed and disposed of or destroyed by prescribed method listed below.

Maintain an accurate count of each type (variety, etc.) of plant bagged for

destruction and record this information.

The use of plastic bags may not be reasonable for the destruction of very large inventories. Other methods such a dumpsters with double layers of plastic liner that can be folded over the top and sealed to prevent debris from escaping during transport or storage may also be used. Contact your regional office or PPQ headquaters for additional guidance.

All bench areas where plants were held, and other areas at risk for exposure to infected plant material, will be disinfected according to prescribed disinfection procedures.

Nursery Plant Disposal and Destruction Methods

Incineration and steam sterilization are the preferred methods of disposal, however, these options may not be practical for large amounts of waste. In these cases, approved landfills, as described below are an option.

1. ***Burning or Incineration:***

All plant material and media must be incinerated or burned to the point of ash. Plastic pots are sometimes not accepted at incinerators and can be removed and sent to a landfill or properly disinfected.

2. ***Steam sterilization under pressure***:

Follow the guidelines in the PPQ Treatment Manual, pages 3.4.1-3.4.2. This is the same as autoclaving.

3. ***Approved Landfill***:

An approved landfill is a State licensed municipal or private facility that is managed under state regulation to meet conditions that would prevent potential pollutants from leaching into groundwater.

See Appendix 10 for a USDA Memorandum for Record concerning landfills that may be used to alleviate concerns of local officials, landfill operators, or the public.

PLEASE NOTE: This plant pathogen is not considered a hazardous waste, biomedical waste, epizootic, or any other substance of concern for human health --*it is an agricultural pathogen only.* Proper disposal is required because there is a remote possibility that it could reach ground water for eventual uptake from wells used for irrigation of host crops. The concentration of the bacteria will be minimal relative to the volume of material disposed of, so the risk is low if the landfill has normal safeguards in place.

The regulatory official that witnesses the disposal of double bagged material from nurseries that use this option for disposal must assure the material is buried under two or more feet of soil. It is suggested that the landfill be notified in advance to arrange for a hole to be dug in the landfill in which the material can be disposed and then covered with soil. If digging a hole is impractical for a landfill operation, another option is to cover the plant material with "alternative daily cover" or composted green waste normally used to cover refuse at landfills. These operations must be witnessed, supervised, and documented by a plant regulatory official.

Nursery Disinfection Procedures

All potentially contaminated surfaces (benches, flats, walkways, footbaths, drainage areas under benches) and equipment within an infected greenhouse or area in contact with infected material will be cleaned of any soil or media, and sprayed and soaked to the point of runoff with approved disinfectants or by pressurized steam cleaning to raise the surface temperature to 212^{o}F. (see PPQ Treatment Manual, page 3.4.2).

Disinfection of Tools and Equipment

Tools and equipment that may have come in contact with infected plants or contaminated soil will be cleaned of any soil or media and disinfected by approved disinfectants applied to the point of runoff or by thoroughly washing with pressurized steam to raise the temperature to 212^{o}F and applied to the point of runoff. (see PPQ Treatment Manual, page 3.4.2).

Exposed Irrigation Systems

A recirculating irrigation system, subirrigation system, or an irrigation system that does not prevent backflow of water from infected greenhouses must be drained and all parts, sumps, and pumps cleaned with approved disinfectant solutions. This also includes sumps, pumps, and recirculating holding ponds. There are disinfectants labeled for disinfecting exposed irrigation systems and there are also acceptable water pre-treatment systems that are employed by large commercial greenhouse operations. These systems, to be effective against *Ralstonia solanacearum* race 3 biovar 2 must have ozonation with 0.4 ppm residual O_3 for 4 minutes with UV light of at least 300 j/m^2 at >50% transmission.

Holding Pond Decontamination Procedures

When holding ponds have become contaminated from irrigation system runoff, contact your regional office and state about possible environmental considerations and treatment options.

Ground Decontamination Procedures

When outdoor soil or holding areas have become contaminated during plant storage or runoff, contact your regions and state about possible environmental consideration and treatment options.

Approved Disinfectants

For disinfection of all tools, benches, walkways, surfaces in contact with potentially infected plant material after removal of soil or media:

Several quaternary ammonia (20% ammonium chloride) products are

approved for greenhouse use. Assure that the label specifies *Ralstonia* or *Pseudomonas* (a previous name for *Ralstonia*) and follow the rates listed on the label for disinfection of surfaces, tools, equipment, benches, walkways, gravel beds under benches, etc. (Check with state labeling requirements as some states may have more stringent or specific disinfection compounds approved for their state).

Some compounds that are registered for *Pseudomonas* are:

Physan® 20 (which is not approved for greenhouse where food crops are grown and not approved for use in California)

Green Shield® (approved for most uses: has a special formulation, Green Shield®CA for use in California).

Maquat® 615-HD or 615-LR. Use rates as directed on the label.

ZeroTol® (27% hydrogen dioxide) is a compound that is not a quaternary ammonia, and has rates for disinfection including surfaces with soil or media contamination that cannot be cleaned. This product is also approved for use in disinfection of contaminated irrigation systems. Use rates as directed on the label.

Consult Appendix 9 for more information on disinfectants.

DISCLAIMER: Any product named in this document does not constitute an endorsement by USDA of that product, but only as one that USDA is aware of that meets specifications or labeling requirements for the use intended. If other products are efficacious and labeling is appropriate, that product can also be used. Please check with your regional or headquarters contact for other product recommendations.

Control Records

Also attach any documentation, receipts, etc. that document these actions. Program personnel must maintain records and maps noting the locations of all detections, the number and type plants subjected to control actions, and the materials and formulations used in each treated area. Attach all documentation to the office EAN copy.

Environmental Monitoring

Contact the PPQ headquaters Environmental Monitoring Staff for guidance on environmental documentation and monitoring.

DEFINITIONS

Approved Landfill	A State licensed municipal or private landfill that is managed under state regulation to meet conditions that would prevent leaching into groundwater of potential pollutants.
Chlorosis	Yellowing of normally green tissue due to chlorophyll destruction in infected plants.
Decontamination	The application of an approved chemical or other treatment to contaminated implements, material, or buildings for killing or deactivating a pathogen.
Detection Survey	A survey conducted in an environmentally favorable area where *R. solanacearum* race 3 biovar 2 is not known to occur.
Destination nursery	Any nursery receiving suspect geraniums or suspect plants, from a rooting station or other nursery.
Disposal	A method used to eliminate diseased plant material or material associated with diseased plant material, usually at an approved landfill.
ELISA	An acronym for Enzyme Linked Immunosorbent Assay, which is a serological laboratory technique used to determine the genus and species in a Ralstonia host sampling and testing program, but not race and biovar.
Host	A plant which is invaded by a parasite or pathogen and from which it obtains its nutrients.
Incineration	Any burning of geranium plants and associated soil or media that results in their complete destruction.
Infection	The establishment of a parasite on or within a host plant.
Monitoring or Evaluation Survey	A survey conducted at a site where a disease was found and where an eradication program is being performed.
Necrosis	Dead or discolored plant tissue.
Pathogen	Any organism that can incite a disease.
PCR	An acronym for Polymerase Chain Reaction, and laboratory technique that amplifies DNA sequences in order to determine the race and biovar in a Ralstonia host sampling and testing program.

Potentially infected plants within nursery property	Includes all plants under the drip-line of hanging suspect geraniums, all plants on a shared irrigation system with a positive testing suspect geraniums and all plants that may have been infected by positive-testing suspect geraniums through unsanitary greenhouse practices.
Positive testing geraniums	Geraniums in which *Ralstonia solanacearum* race 3 biovar 2 has been confirmed to be present by the USDA-PPQ-CPHST laboratory in Beltsville, MD.
Shipment	All plants that were prepared for shipment and transported with suspect geraniums from a common supplier, i.e., listed on the invoice or packing list.
Suspect geraniums	Geraniums (*Pelargonium* spp) that are associated with positive testing geraniums. These may have originated from a foreign country, a rooting station, or another nursery and be the same variety or in the same shipment depending on the circumstances.
Symptom	The external and internal reactions or alterations of a plant as the result of a disease.
Traceback	To investigate the origin of infestated plants through intermediate steps in commercial distribution channels to the origin.
Traceforward	To investigate where infected plants may have been distributed from a source through steps in commercial distribution channels.

Appendix 1
Questionnaire for Nursery Owner/ Manager

Name of Nursery _____ Name of Owner _____

Address of Nursery _____

(city)_____(state)_____ (zip code)_____

Phone number and contact name, title _____
GPS coordinates if available_____

Type of nursery facility (circle) Rooting Station Direct Ship Wholesaler

Other _____ Wholesaler Retailer

1) Indicate the and numbers of suspect geranium plants by variety and shipping date received from the foreign facility, rooting station, or wholesaler. (Indicate the name of the shipper you received the plants from).

2) The current location and numbers of all suspect geranium varieties or shipments in question.

3) The history of movement of suspect geraniums within the facility.

4) The history of movement of suspect geraniums out of the facility. If shipped out, to what customers, what quantity of each variety, and the dates shipped.

5) The condition of plants (observations of wilting) since received. (numbers and variety showing symptoms).

6) If symptoms were observed this season, where within the facility were the wilting plants noticed and which variety?

7) If you had dead or wilted plants, what was the disposition of dead or culled plants and soil/potting media associated with those plants? If they were disposed of, where?

8) What was the location and number of plants of other hosts (tomato, eggplant, potato, peppers) in your facility inventory since receiving suspect shipments?

9) What type of irrigation system do you use? (i.e., sub-irrigation, ebb and flow, drip, hand watering, etc.) Is there backflow prevention in place?

10) If flood, sub-irrigation, or ebb and flow, is practiced, identify the location of all plants sharing the same water source with suspect geraniums.

11) Do you filter or treat your irrigation water? Do you use water from an outdoor holding pond for irrigation recirculation or overflow?

12) Describe the type of greenhouse benches used and floor composition.

13) Do you ever store plants on the ground? If so, on what kind of surface?

14) Do you keep you varieties or shipment segregated?

15) Do you use a particular tagging or tracking system for geranium plants you distribute? If so, please describe the system used.

16) Do you perform propagation of geraniums from the plants you receive from foreign sources?

17) What kind of protection do your nursery workers use when handling plants? How often do they wash their hands or cutting implements?

18) Do you use a disinfectant ? If so, what is the name of the disinfectant?

19) Is there a standard greenhouse sanitation protocol document available for your facility? Please provide a copy if so.

The answers to these questions will help agricultural officials determine the extent of possible contamination in your nursery facility. Thank you for your cooperation.

_____ _____

owner or manager signature *date*

PPQ form 391	

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.

FORM APPROVED
OMB NO. 0579-0010

U.S. DEPARTMENT OF AGRICULTURE
ANIMAL AND PLANT HEALTH INSPECTOIN 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 USDA USE
LOT NO.
PRIORITY

1. COLLECTION NUMBER

2. DATE MO | DA | YR

3. SUBMITTING AGENCY ☐ State Cooperator ☐ PPQ ☐ Other .....

4. NAME OF SENDER

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

6. ADDRESS OF SENDER

INTERCEPTION SITE

7. NAME AND ADDRESS OF PROPERTY OR OWNER

ZIP

COUNTRY/ COUNTY

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

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

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

10. HOST INFORMATION

NAME OF HOST *(Scientific name when possible)*

11. QUANTITY OF HOST

NUMBER OF ACRES/PLANTS

PLANTS AFFECTED *(Insert figure & indicate number or percent)* ☐ Number ☐ Percent

12. PLANT DISTRIBUTION

☐ LIMITED
☐ SCATTERED
☐ WIDESPREAD

13. PLANT PARTS AFFECTED

☐ Leaves, Upper Surface
☐ Leaves, Lower Surface
☐ Petiole
☐ Stem
☐ Trunk/Bark
☐ Branches
☐ Growing Tips
☐ Roots
☐ Bulbs, Tubers, Corms
☐ Buds
☐ Flowers
☐ Fruits or Nuts
☐ Seeds

14. PEST DISTRIBUTION

☐ FEW
☐ COMMON
☐ ABUNDANT
☐ EXTREME

15. ☐ INSECTS ☐ NEMATODES ☐ MOLLUSKS

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

16. SAMPLING METHOD

17. TYPE OF TRAP AND LURE

18. TRAP NUMBER

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

23. TENTATIVE DETERMINATION

24. DETERMINATION AND NOTES *(Not for Field Use)*

FOR USDA USE
DATE RECEIVED
NO.
LABEL
SORTED
PREPARED
DATE ACCEPTED
RR

PPQ FORM 391
(JUL 86)

Previous editions are obsolete.

Sample Accompaniment form	*To be placed in the outside bag of the double bag plant or culture sample in addition to PPQ form 391)*

Ralstonia Suspect Sample Accompaniment Form

(To be placed in the outside bag of the double bag plant or culture sample in addition to PPQ form 391)

Date sample taken _____ Date sample submitted _____

Assigned Sample Number ____-_____-_____ (*state code-facility code-00000*)

Name of Nursery and contact name _____

Location of Nursery _____City _____State ___

Phone # _____ E-mail address _____

Sample representing shipment invoice #_____ week shipped _____

Shipper company name _____ origin if known_____

Specific number and type of plants in the shipment_____

_____

Location of shipment in nursery _____

include greenhouse and bench number(s), section, etc.

Complete name of plant _____

genus, species, cultivar/variety, color, any other descriptor words such as series, class

Name, agency and phone number of person taking the sample

_____

Lab name, phone number of lab, and name of contact person performing screening test

_____

Ralstonia Suspect Sample Accompaniment Form

(To be placed in the outside bag of the double bag plant or culture sample in addition to PPQ form 391)

Date sample taken _____ Date sample submitted _____

Assigned Sample Number ____-_____-_____ (*state code-facility code-00000*)

Name of Nursery and contact name _____

Location of Nursery _____City _____State ___

Phone # _____ E-mail address _____

Sample representing shipment invoice #_____ week shipped _____

Shipper company name _____ origin if known_____

Specific number and type of plants in the shipment_____

_____

Location of shipment in nursery _____

include greenhouse and bench number(s), section, etc.

Complete name of plant _____

genus, species, cultivar/variety, color, any other descriptor words such as series, class

Name, agency and phone number of person taking the sample

_____

Lab name, phone number of lab, and name of contact person performing screening test

_____

Appendix 3 DIAGNOSTIC VALIDATIONS AND ISOLATION METHODS

Laboratories requiring the information in this appendix should contact their State Plant Health Director or State Plant Regulatory Official.

Appendix 4 State Screening Diagnostic Laboratories

The following laboratories are designated as screening diagnostic laboratories for making determination to the genus and species only. Laboratories making positive determinations to this level must notify their State Plant Regulatory Official (SPRO) and State Plant Health Director (SPHD) when sending samples for race and biovar determination. SPRO contacts are at:http://www.aphis.usda.gov/npb/npbmemb.html#Membership%20Dirctory SPHD contacts are at: http://www.ceris.purdue.edu/napis/names/sphdXstate.html

Laboratories receiving samples from within their own states do not need USDA Plant Pest Permits, but laboratories receiving samples from other states do require one. Information and forms for acquiring a Plant Pest Permit (PPQ form 526) can be found at: *http://www.aphis.usda.gov/ppq/permits/plantpest/index.html* For information on the laboratories in the National Plant Diagnostic Network, see the last pages of this section.

Further determination to the race and biovar requires additional registrations for Select Agents under the Agriculture Bioterrorism Protection Act of 2002. Currently, the only laboratory with proper registrtations to make these determinations for *Ralstonia solanacearum* race 3 biovar 2 is the USDA, APHIS, PPQ National Plant Germplasm and Biotechnology Laboratory in Beltsville, MD. For more information regarding select agent permitting, see: *http://www.aphis.usda.gov/ppq/permits/agr_bioterrorism/*
For information on packaging and documentation, see the Diagnostics section.

State Screening Diagnostic Laboratories

Alabama
Contact: Dr. Jackie Mullen
Extension Plant Pathologist
Plant Diagnostic Lab
Auburn University
Auburn, AL 36849
Phone: 334-844-5508
jmullen@acesag.auburn.edu

Alaska
Not yet determined.

Arizona Will use CDFA*:
Contact: Dr. Dan Opgenorth
California Department of Food and Agriculture
Plant Pest Diagnostics Laboratory
3294 Meadowview Road
Sacramento, CA 95832-1448
Phone (916) 262-1100 1100
Fax (916) 262- 1190 E-mail: DOpgenor@cdfa.ca.gov

Arkansas
Contact: Dr. Stephan Vann
University of Arkansas Cooperative Extension Service
Plant Diagnostic Clinic
2001 Hwy 70 East
Lonoke, AR 72086
Phone (501) 676-3124

California
Contact: Dr. Dan Opgenorth
California Department of Food and Agriculture
Plant Pest Diagnostics Laboratory
3294 Meadowview Road
Sacramento, CA 95832-1448
Phone (916) 262-1100 1100
Fax (916) 262- 1190
E-mail: DOpgenor@cdfa.ca.gov

Colorado
Contact: Tamela Blunt
Center for Crop Biosecurity,
Colorado State University
FT. Collins, CO 80526
Phone: 970-491-6950

Connecticut
Contact: Dr. Sharon Douglas
CT Agricultural Experiment Station
POB 1106
New Haven, CT 06504
Phone: 203 974-8499
Sharon.douglas@po.state.ct.us

Delaware
Contact: Dr. Bob Mulrooney
Department of Plant and Soil Sciences
Room 152 Townsend Hall
University of Delaware
Newark, DE 19717
Phone: 302-831-4865
Fax: 302-831-0605
bobmul@udel.edu

District of Columbia
Not yet determined.

Florida
Contact: Dr. Tim Schubert FDACS-DPI
P O Box 147100
Gainesville, FL 37614-7100
Phone: 352-372-3505 x 143
schubet@doacs.state.fl.us

Georgia
Contact: Dr. Jean Woodward
University of Georgia
Plant Pathology Department
2106 Miller Plant Science Bldg.
Athens, GA 30602-7274
Phone: (706) 542-9146
jwoodwar@uga.edu

Hawaii
Not yet determined.

Idaho
Contact: Liz Vavricka
Plant Industry Lab
Idaho State Department of Agriculture
PO Box 790
Boise, ID 83701
Phone: 208-332-8640

Illinois
Contact: Mike Tiffany
*Agdia, Inc.
30380 County Road 6
Elkhart, IN 46514
Phone: 574.264.2014
info@agdia.com

Indiana
Contact: Dr. Karen Rane
Plant Pest Diagnostic Lab
Purdue University
915 W. State Street
West Lafayette, IN 47907-2054
Phone: 765.494.5821
rane@purdue.edu

Iowa
Contact: Paula Flynn
ISU Plant Disease Clinic
323 Bessey Hall
Iowa State University
Ames, Iowa 50011
Phone: 515-294-0581

Kansas
Dr. Ned Tisserat,
Plant Pathology Department
Kansas State University
Manhattan, Kansas
telephone number is 785-532-1387.

Kentucky
Dr. John Hartman S-305 Ag Sci Bldg N
University of KY
Lexington, KY 40546
Phone: 859 257-5779
jhartman@ca.uky.edu

Lousiana
Contact: Dr. Clayton A. Hollier
LSU AgCenter
Baton Rouge, La.
Phone: 225-578-2186
Specialist, Plant Pathology LSU AgCenter
Other contacts at Dept. Plant Pathology - LSU: (Dr. Chris Clark, 225-578-1381, and Dr. Gordon Holcomb 225-578-1386)
State of Louisiana
Contact: Craig Roussel or Tad Hardy
Louisiana Dept. Agriculture and Forestry
Baton Rouge, La.
Phone: 225-952-8100

Maine
Contact: Bruce Watt
University of Maine
Pest Management Office
491 College Avenue
Orono, ME 04473-1295
phone (207) 581-3880
bwatt@umext.maine.edu

Maryland
Contact: Jennifer Dominiak
Maryland Department of Agriculture
50 Harry S. Truman, Pkwy.,
Room 345
Annapolis, Maryland 21401
Phone: 410-841-5920
dominijd@mda.state.md.us

Massachusetts
Contact: Dr. Robert L. Wick
Department of Microbiology
Morrill Science Center N203
University of Massachusetts
Amherst, MA 01003
Phone: 413-545-1045
rwick@pltpath.umass.edu

Michigan
Contact: Dr. Richard Kaitany
Geagley Laboratory
Michigan Department of Agriculture
1615 S. Harrison Rd.
East Lansing, MI 48823
Phone: (517) 337-5091
kaitanyr@msu.edu

Minnesota
Contact: Gary Horvath
Minnesota Department of Agriculture
Laboratory Services Division
90 W. Plato Blvd.
St. Paul, MN 55107-2094
Phone: 651.215.9063
Fax: 651.297.8787
gary.horvath@state.mn.us

Mississippi
Contact: Dr. Alan Henn
Associate Extension Professor/Entomology and Plant Pathology
Mail Stop 9655
Mississippi State, MS 39762
Phone: 662.325.4535
ahenn@ext.msstate.edu

Missouri
Contact: Dave Johnson, Plant Pathologist
Missouri Department of Agriculture
Plant Industries Division
115 Constitution Drive
P.O. Box 630
Jefferson City, Missouri 65102
Phone: (573) 751-8319
FAX: (573) 526-7777

Montana
Contacts: Martha Mikkelson/Barry Jacobsen
Plant Disease Clinic
119 Ag BioScience Facility
Montana State University-Bozeman
Bozeman, MT 59717-3150
Phone: (406)994-5150

Nebraska
Contact: Jennifer Chaky
Plant Pathology Department,
448 Plant Science Building,
University of Nebraska, Lincoln,
Lincoln NE 68583
Phone: 402-472-8725

Nevada
Contact: Shouhua Wang
Plant Pathologist
Nevada Department of Agriculture
Plant Division
350 Capitol Hill Avenue
Reno, NV 89502
Phone: (775)688-1182x246
Fax: (775)688-1178
Email: shwang@govmail.state.nv.us

New Hampshire
Contact: Cheryl Smith
Extension Education / Adjunct Professor
University of New Hampshire
Spaulding Life Sciences Bldg., Room 242
Durham, NH 03824-3544
Phone: 603.862.3841
Cheryl.smith@unh.edu

New Jersey
Dr. Glenn Freeman
New Jersey Department of Agriculture
Plant Laboratory Services
PO Box 330
Trenton, NJ 08625-0330
609.292.5484
glenn.freeman@ag.state.nj.us

New Mexico
Contact: Sherry Sanderson Bureau Chief.
NMDA Bureau Chief
Entomology and Nursery Industries, MSC 3 BA,
Las Cruces, NM 88003-0005.
Phone 505-646-3207

New York
Contact: Margery Daughtrey
Cornell University
Long Island Horticultural Research & Extension Center
3059 Sound Ave.
Riverhead, NY 11901
Phone: 631.727.3595
mld9@cornell.edu

Contact: Karen Snover-Clift
Cornell University
Department of Plant Pathology
334 Plant Science Bldg.
Ithaca, NY 14853
Phone: 607.255.7850
kls13@cornell.edu

North Carolina
Tom Creswell
NCSU Plant Disease and Insect Clinic
William Hall, Room 1104
100 Derieux Hall
Campus Box 7211
Raleigh, NC 27695-7211
Phone: 919.515.3619
Fax: 919.515.7716
tom_creswell@ncsu.edu

North Dakota
Contact: Mike Tiffany
*Agdia, Inc.
30380 County Road 6
Elkhart, IN 46514
Phone: 574.264.2014

Ohio
Contact: Nancy J. Taylor
Extension Associate & Coordinator
Plant Pest Diagnostic Clinic
110 Kottman Hall
2021 Coffey Road
Columbus, Ohio 43210-1087
Phone: 614-292-5006
Fax: 614-292-7162
Internet: taylor.8@osu.edu

Oklahoma
Contact: Dr. Brian Olson
Oklahoma State University
Ento. and Plant Pathology
119 NRC
Stillwater, OK 74078-3033
Phone: (405) 744-7126
olsonb@okstate.edu

Oregon
Contact: John A. Griesbach, Ph.D.
Acting Supervisor
Plant Health Lab
Plant Health Section
Oregon Dept. of Agriculture
635 Capitol St. NE
Salem, OR 97301-2532
jgriesba@oda.state.or.us
Phone: (503) 986-4636 or (503)986-4661
Fax: (503) 986-4786

Pennsylvania
Contact: Dr. Seong Kim
Pennsylvania Dept. of Agriculture
Bureau of Plant Industry
Plant Pathology Lab
2301 North Cameron Street
Harrisburg, PA 17110-9408
Phone: 717.772.5221
Fax: 717.705.6518
skim@state.pa.us

Rhode Island
Contact: Mike Tiffany
*Agdia, Inc.
30380 County Road 6
Elkhart, IN
Phone: 574.264.2014
info@agdia.com

South Carolina
Contact: Mike Tiffany
Agdia, Inc.
30380 County Road 6
Elkhart, IN 46514
Phone: 574.264.2014
info@agdia.com
and

Contact: Meg Williamson
Clemson University Plant Problem Clinic
171 Old Cherry Road
Clemson, SC 29634-0114
Phone: (864) 656-3125
ppclnc@clemson.edu

South Dakota
Contact: Dr. Marty Draper
South Dakota State University
Brookings ,SD 57006
Phone: (605) 688-5157

Tennessee
Contact: Mike Tiffany
*Agdia, Inc.
30380 County Road 6
Elkhart, IN 46514
Phone: 574.264.2014
info@agdia.com

Texas
Contact: Dr. Larry Barnes
Texas Plant Disease Diagnostic Laboratory
1500 Research Parkway, Suite A-130
College Station, Texas 77845
Telephone (979)845-8032
Email l-barnes@tamu.edu

Utah
Contact: Scott C. Ockey
Plant Disease Diagnostician/Senior Research Associate
Utah State University
Old Main Hill
Logan, UT 84322-5305
Phone (435)797-2435
Fax (435)797-1575
scotto@ext.usu.edu

Virginia
Contact: Grace O'Keefe
Washington Building, Room 703
1100 Bank Street
Richmond, VA 23219
Phone: 804.786.3515
804.371.7793 – fax
gokeefe@vdacs.state.va.us

Vermont
Contact: Scott Pfister
Vermont State Plant Pathologist
103 S. Main Street
Waterbury, VT 05671-0101
Phone: 802.828.3481
spfister@agr.state.vt.us

Washington
Contact: Art Wagner, Plant Pathology Project Coordinator
Plant Pathology Laboratory
3939 Cleveland Avenue S.E.
Olympia, WA 978501-4079
Fax Number: (360) 586-5286
awagner@agr.wa.gov

West Virginia
Contact: Gary Gibson
WVDA, Plant Industries Division
1900 Kanawha Blvd., East
Charleston, WV 25305-0191
Phone: 304.558.2212
ggibson@ag.state.wv.us

Wisconsin
Contact: Anette Phibbs
WDATCP, ARM, Plant Industry Division
4702 University Avenue
Madison, WI 53705
Phone: 608.266.7132
Fax: 608.266.5855
anette.phibbs@datcp.state.wi.us

Wyoming
Contact: Mr. Gary Franc
University of Wyoming
Dept. of Plant Pathology
P.O. Box 3354
Laramie, Wyoming 82071-3354
Phone: (307) 766-2397

***** *States sending samples for Ralstonia solanacearum determination to other states must assure that the laboratory receiving samples has a Plant Pest Permit (PPQ form 526) to receive those samples from out-of-state. For more information, see:*
http://www.aphis.usda.gov/ppq/permits/plantpest/index.html

The National Plant Diagnostic Network (NPDN)

Diagnostic screening can also take place at a laboratory designated in the CSREES National Plant Diagnostic Network divided into five regions. If using the laboratory in your region and in another state, these laboratories have the proper USDA Plant Pest Permit to receive diagnostic plant samples from out of state. They can make determinations to the genus and species level, but are not yet under permits to do race and biovar testing.

Northeast Plant Diagnostic Network (NEPDN): the Northeast region serves the following states; Connecticut, Delaware, Maine, Maryland, Massachusetts, New Hampshire, New Jersey, New York, Pennsylvania, Rhode Island, Vermont, and West Virginia.

NEPDN, Cornell University
Karen L. Snover-Clift
Department of Plant Pathology
334 Plant Science Bldg.
Ithaca, NY 14853
kls13@cornell.edu
607-255-7850
Fax-607-255-4471

Great Plains Diagnostic Network (GPDN): the Great Plains region serves the following states; Colorado, Kansas, Montana, North Dakota, Oklahoma, South Dakota, northern Texas, Nebraska, and Wyoming.

GPDN, Kansas State University
Ned Tisserat
Department of Plant Pathology
4024 Throckmorton Hall
Manhattan, KS 66506
tissne@ksu.edu
(785) 532-1383

North Central Plant Diagnostic Network (NCPDN): the North Central region serves the following states; Iowa, Michigan, Ohio, Indiana, Illinois, Minnesota, Missouri, and Wisconsin.

NCPDN, Michigan State University
Jan Bryne
114 CIPS
Michigan State University
East Lansing MI 48824-1311
byrnejm@msu.edu
(517) 355-3504

Southern Plant Diagnostic Network (SPDN): the Southern region serves the following states; Alabama, Louisiana, southern Texas, Arkansas, Mississippi, Virginia, Florida, North Carolina, Puerto Rico, Georgia, South Carolina, Kentucky, and Tennessee.

SPDN, University of Florida
Richard Cullen
Plant Disease Clinic
UF Bldg 78 Mowry Road
PO Box 110830
Gainesville FL 32611-0830
(352) 392-1795
(352) 392-3631 ext 254 (Carrie Harmon)

Western Plant Diagnostic Network (WPDN): the Western region serves the following states; American Samoa, Arizona, California, Hawaii, Idaho, Alaska, Nevada, New Mexico, Oregon, Guam, Utah, and Washington.

WPDN, Oregon State University
Melodie Putnam
1089 Cordley Hall
Corvallis, OR 97331-2903
(541) 737-3472
putnamm@science.oregonstate.edu

Private Laboratory Approved for Diagnostic Screening:
There is one private laboratory that has permits and necessary procedures in place to receive plant samples from out-of-state for diagnostic screening to genus and species:

Agdia Inc.,
30380 County Road 6,
Elkhart, IN 46514
phone number: (574) 264-2014, or 1-800-62-AGDIA
www.agdia.com

Other private laboratories may be eligible as permits are approved and these establishment names will be provided.

Appendix 5 Inspectors are to use the following Control Action Sheets to document the actions taken at nursery facilities. One is the locations in the Eastern Region and the other is for the Western Region. Provide as much detail and documentation as appropriate to document actions.

PPQ ER Control Action Information Sheet for *Ralstonia solanacearum race 3 biovar 2*

Please write legibly or type.

Before undertaking control actions:

- **Fax a copy of this form to Billy Newton & Lloyd Garcia @ 919-716-5656.**
- **Fax a copy of all EAN's/Stop Sale Orders associated with this greenhouse.**
- **Fax a copy of the PPQ 391 for the site.**
- **E-mail digital photos of the site.**

State:

Name & address of greenhouse:

Latitude (in decimal degrees)	Longitude (in decimal degrees)

Date of *Rs r3b2* confirmation from PPQ Beltsville, MD	
Confirmation number	
Number of positive greenhouses	
Number of associated greenhouses	
Type of irrigation system	
Proposed method of destruction	
Proposed method of greenhouse/irrigation system disinfection	

Numbers, Species & Varieties of plants Destroyed

Species	Variety	Number Destroyed

Chronological narrative & sketch of location:

PPQ WR Control Action Work Sheet

Establishments positive for *Ralstonia solanacearum r3b2*

Please write legibly or type.
Keep your Regional contact informed of Control Actions

- **Necessary actions are:**
- **Obtain copies of all EAN's or stop sale orders associated with this greenhouse.**
- **Obtain a copy of the PPQ 391 for the site.**
- **For difficult control problems take digital photos of the site.**

- **If you have questions call Cliff Smith @ 970-494-7568.**

State:

Name & Address of Greenhouse:

Date of Ralstonia Confirmation from PPQ Beltsville, MD	
Confirmation Number	
Number of Positive Greenhouses	
Number of Associated Greenhouses	
Type of Irrigation System	
Proposed Method of Destruction	
Proposed Method of Greenhouse/Irrigation Disinfection	

PPQ WR Control Action Work Sheet

Establishments positive for *Ralstonia solanacearum r3b2*

Numbers & Species of Plants

Species	Number

Chronological narrative of events & sketch of location:

Appendix 6

GERANIUMS IN THE NURSERY TRADE

Common *Pelargonium* species in the nursery trade

Geraniums is a common name that can refer to plant species in the genus *Pelargonium* and the genus *Geranium.* The common nursery plant sold is one of several *Pelargonium* species listed below. The Ralstonia program does not regulate plants in the genus *Geranium*, which are primarily perinneal landscape plants not know to be hosts of *R. solanacearum*.

Pelargonium xhortorum: Zonal geranium or Florist's geranium. "Americana" geraniums fall in this group. This genus and species makes up 70-80% of the geraniums sold in the U.S. each year. Vegetatively propagated and most are tetraploids.

Pelargonium xhortorum: Seed geraniums. A smaller portion of the market. These are diploids and are reproduced via seed

Pelargonium peltatum: Ivy geranium. Approx 10-20% of the market is this species. Vegetatively propagated. Typically grown mainly in baskets (hanging above other crops). There are also seed propagated lines of this species.

Pelargonium domesticum: Regal or Martha Washington geraniums. This species is grown mainly as a flowering potted crop through florists and upper end retail garden centers. Vegetatively propagated. No more than 5-10% of the overall market is of this species. These are used as pot plants to display in the home and typically are planted out into the garden since they will not flower during the heat of the summer. The foliage and flowers are significantly different than the zonal geraniums.

Pelargonium spp.: Scented geraniums and other novelty types with unusual flowers, foliage or scented foliage. Very small market segment of unusual types. Vegetatively propagated. Broad diversity of genetics make up this group and difficult to type to species.

Appendix 7 Symptomology on Hosts

Symptoms of *R. solanacearum* race 3, biovar 2 on geraniums

Figure 1. Early wilting symptoms caused by infection with *R. solanacearum* race 3, biovar 2. *Photo courtesy of the Wisconsin Department of Agriculture, Trade and Consumer Protection*

Figure 2. More advance wilting and abnormal leaf yellowing symptoms (chlorosis) caused by infection with *R. solanacearum* race 3, biovar 2. *Photo courtesy of the Wisconsin Department of Agriculture, Trade and Consumer Protection*

Figure 3. Abnormal yellowing symptoms caused by infection with *R. solanacearum* race 3, biovar 2. *Photo courtesy of the Plant Disease Diagnostics Clinic, University of Wisconsin-Madison/Extension*

Figure 4. Close-up of wilting and necrosis caused by infection with *R. solanacearum* race 3, biovar 2. *Photo courtesy of Margery Daughtrey, Cornell University*

Figure 5. Wilting and mortality of geraniums caused by infection with *R. solanacearum* race 3, biovar 2. *File photo*

Figure 6. One plant showing mortality by *R. solanacearum* race 3, biovar 2 and another showing early wilt symptoms caused by infection with *R. solanacearum* race 3, biovar 2. *File photo*

Symptoms of Bacterial Blight caused by *Xanthomonas pelargonii* on geranium

Figure 7. Wilting symptons caused by Bacterial Blight, *Xanthomonas pelargonii*, are indistinguishable from *Ralstonia solanacearum* wilting. *Photo courtesy of Margery Daughtrey, Cornell University*

Figure 8. Bacterial Blight, *Xanthomonas pelargonii*, also causes characteristic spotting of leaf tissue *Photo courtesy of Margery Daughtrey, Cornell University*

Appendix 8

Emergency Action Notification Instructions

Emergency Action Notification and its Use in Geranium Holds

The Emergency Action Notification (PPQ form 523) is used to hold suspect geraniums and potentially infected plants at nurseries or other facilities. Only one form should be used to hold each lot of suspect geraniums and associated potentially infected plant material. The same form will be used for determine what is held, what is ordered destroyed, and what is released.

Forms can be filled out by hand or using the electronic version in PPQ Lotus Notes. Follow the instructions below.

Emergency Action Notification Procedures

For Block 1, enter the name of location of the nearst PPQ office. Under "Name of Article" in block 3, enter *Pelargonium* spp.", not "Geranium". In Block 4 ,enter the greenhouse numbers or other information indicating the location of the plants held. In the Shipper Block 6, enter the source nursery or foreign country shipper. Blocks 7 and 8 can be left blank unless that information is known.

To place plants within a nursery on "Hold" , in Block 12 of the EAN, enter for the Pest : "*Ralstonia solanacearum* race 3 biovar 2". The authority underwhich actions are taken is 7 CFR 330 and the Plant Protection Act. In block 15, the Action Required text is as follows:

All geranium (*Pelargonium* spp.) _____ varieties received from _____ during the dates _____ are prohibited from movement from the nursery property pending further notification by USDA APHIS PPQ. Any other plant material received by those same shipments that may have been exposed directly, in shipping or since being received, by shared irrigation systems, or by unsanitary nursery cultural practices are also subject to this hold. All host plants associated with the above exhibiting symptoms of wilt must be reported immediately to USDA APHIS PPQ and held until further notice. No other potential host material of *Ralstonia solanacearum* race 3 biovar 2 may leave greenhouses containing suspect plant material until further evaluations can be made. The above listed plants and all potentially infested material after further investigations are conducted will be destroyed either by incineration, steam sterilization, or an approved landfill in accordance with USDA policies. Areas housing infected material shall be disinfected according to USDA protocols.

Releasing Plants

After all hosts have been held at a nursery with suspect geraniums and investigations have determined which other plants are potentially infected because of shared water or unsanitary nursery practices, release all other plants. Make a notation in block 16.

Documenting Actions Taken

In block 19, after plants have been destroyed and area sanitized, indicate so with the method used to destroy the plants, i.e.,

"Incineration", "Steam Sterilization", or "Disposal at Approved Landfill" and the name of the disinfectant(s) used including what articles were disinfected.

U.S. DEPARTMENT OF AGRICULTURE
ANIMAL AND PLANT HEALTH INSPECTION SERVICE
PLANT PROTECTION AND QUARANTINE

EMERGENCY ACTION NOTIFICATION

SERIAL NO.

1. PPQ LOCATION

2. DATE ISSUED

3. NAME AND QUANTITY OF ARTICLE(S)

4. LOCATION OF ARTICLES

5. DESTINATION OF ARTICLES

,

6. SHIPPER

7. NAME OF CARRIER

8. SHIPMENT ID NO.(S)

9. OWNER/CONSIGNEE OF ARTICLES

Name

Address:

,

UNITED STATES

PHONE NO. FAX NO.

SS NO. TAX ID NO.

10. PORT OF LADING

,

11. DATE OF ARRIVAL

12. ID OF PEST(S)

12a. PEST ID NO.

12b. DATE INTERCEPTED

13. COUNTRY OF ORIGIN

14. GROWER NO.

15. FOREIGN PHYTOSANITARY CERTIFICATE NO.

15a. PLACE ISSUED

,

15b. DATE

Under Section 412 and 414 of the Plant Protection Act (7 U.S.C. 7712 and 7714) and Section 2 of the Act of February 2, 1903 (21 U.S.C. 111) and Section 2 of the Act of July 2, 1962 (21 U.S.C. 134a) you are hereby notified, as owner or agent of the owner of said carrier and/or premises and/or articles, to apply remedial measures for an injurious agricultural pest as specified in Item 12, in a manner satisfactory to and under supervision of an Agriculture Officer. Remedial measures shall be in accordance with the action indicated in Item 16 and shall be completed within the specified time indicated in Item 17.*

AFTER RECEIPT OF THIS NOTIFICATION, ARTICLES AND/OR CARRIERS HEREIN DESIGNATED MUST NOT BE MOVED EXCEPT AS DIRECTED BY AN AGRIGULTURE OFFICER. THE LOCAL OFFICER MAY BE CONTACTED AT:

16. ACTION REQUIRED

- ☐ TREATMENT
- ☐ RE-EXPORTATION
- ☐ DESTRUCTION
- ☐ OTHER

* Should the owner or owner's agent fail to comply with this order within the time specified below, USDA is authorized to recover from the owner or agent all costs, including the officer's time, incurred in order to properly dispose of or treat the specified articles.

17. AFTER RECEIPT OF THIS NOTIFICATION COMPLETE SPECIFIED ACTION WITHIN (Specify No. Hours or No. Days):

18. SIGNATURE OF OFFICER

ACKNOWLEDGEMENT OF RECEIPT OF EMERGENCY ACTION NOTIFICATION

I hereby acknowledge receipt of the foregoing notification.

SIGNATURE AND TITLE

DATE AND TIME

19. REVOCATION NOTIFICATION

ACTION TAKEN

SIGNATURE OF OFFICER

DATE

PPQ FORM 523 (JULY 2001) Previous editions are obsolete

Appendix 9 DISINFECTANTS INFORMATION CHART

Table 6. Disinfectants registered against *Pseudomonas** bacteria, with use sites, and other information. Not all of these are listed in the body of the Control section, however, some additional formulations may have uses in sites not approved for those listed. Consult the label to determine appropriate uses.

**Ralstonia solanacearum = Pseudomonas solanacearum*

Trade Name	EPA Reg. No.	Active Ingredients	Equal To	Use Sites	Pertinent Labeled Bacteria	Pertinent unlabeled Bacteria (efficacy data available)
Green Shield	499-368	n-Alkyl dimethyl benzyl ammonium chloride; n-Alkyl dimethyl ethylbenzyl ammonium chloride	Physan 20	Work area, benches, pots, flats, flower buckets, cutting tools, greenhouse glass, walkways, evaporative coolers, decorative pools, fountains, and water displays		*Pseudomonas* spp.
Physan 20	55364-5	n-Alkyl dimethyl benzyl ammonium chloride; n-Alkyl dimethyl ethylbenzyl ammonium chloride	Green Shield	Greenhouses, hard surfaces, lawn and turf grass, seedlings, cut flowers, decorative fountains, pools, birdbaths, and plants	*Pseudomonas* spp.	
Zero Tol	70299-1	Hydrogen dioxide		Greenhouse structures, benches, pots, watering systems, evaporative coolers, storage rooms, ventilation equipment, floors and other equipment	*Pseudomonas* spp.	
Consan Triple Action 20	58044-3	n-Alkyl dimethyl benzyl ammonium chloride; n-Alkyl dimethyl ethylbenzyl ammonium chloride	Triathlon	Greenhouse hard surfaces, work area, benches, flower pots, buckets, flats, cutting tools, walkways, garden bird baths, and evaporative coolers		
Triathlon	58044-3-59807	n-Alkyl dimethyl benzyl ammonium chloride; n-Alkyl dimethyl ethylbenzyl ammonium chloride	Consan Triple Action 20	Greenhouse hard surfaces, work area, benches, cutting tools, walkways, garden bird baths, and evaporative coolers		
Formulation AE-90	47371-89	n-Alkyl dimethyl benzyl ammonium chloride; n-Alkyl dimethyl ethylbenzyl ammonium chloride		Non-porous, inanimate surfaces such as floors, walls, metal surfaces, stainless steel surfaces, plastic surfaces, knobs, handles, and railings	*Pseudomonas aeruginosa*	
Lonza Formulation S-18	6836-77	Octyl decyl dimethyl ammonium chloride; Dioctyl dimethyl		Farm, Poultry, Swine, and Mushroom Premise Sanitation	*Pseudomonas aeruginosa and*	

Trade Name	EPA Reg. No.	Active Ingredients	Equal To	Use Sites	Pertinent Labeled Bacteria	Pertinent unlabeled Bacteria (efficacy data available)
		ammonium chloride; didecyl dimethyl ammonium chloride; n-Alkyl dimethyl benzyl ammonium chloride		Veterinary Practice/Animal Care/Animal Laboratory Disinfection	*Pseudomonas cepacia*	
MAQUAT 128-MT	10324-112	Octyl decyl dimethyl ammonium chloride; Dioctyl dimethyl ammonium chloride; didecyl dimethyl ammonium chloride; n-Alkyl dimethyl benzyl ammonium chloride		Outer clothing, field harvesting equipment, walls/floors of coolers, flower buckets, and greenhouse packing areas	*Pseudomonas aeruginosa*	
MAQUAT 64 MN	10324-113	Octyl decyl dimethyl ammonium chloride; Dioctyl dimethyl ammonium chloride; didecyl dimethyl ammonium chloride; n-Alkyl dimethyl benzyl ammonium chloride		Florist shops, wholesale florist, shippers, greenhouse packing areas, flower buckets, floors/walls of coolers, design and packaging benches, and counter tops	*Pseudomonas aeruginosa*	
MAQUAT 615-HD	10324-72	Octyl decyl dimethyl ammonium chloride; Dioctyl dimethyl ammonium chloride; didecyl dimethyl ammonium chloride; n-Alkyl dimethyl benzyl ammonium chloride	MAQUAT 615-LR	Greenhouses, hard non porous surfaces (flower buckets, floors, walls, coolers, design, packing benches and counter tops)	*Pseudomonas aeruginosa, Xanthomonas axonopodis pv. Citri, and Xanthomonas campestris pv. Vesicatoria*	
MAQUAT 615-LR	10324-109	Octyl decyl dimethyl ammonium chloride; Dioctyl dimethyl ammonium chloride; didecyl dimethyl ammonium chloride; n-Alkyl dimethyl benzyl ammonium chloride	MAQUAT 615-HD	Greenhouses, hard non porous surfaces (flower buckets, floors, walls, coolers, design, packing benches and counter tops)	*Pseudomonas aeruginosa, Xanthomonas axonopodis pv. Citri, and Xanthomonas campestris pv. Vesicatoria*	

[1] Active Ingredient (the total percentage of active ingredients).
[1] Labeled dilution rate for greenhouse versus labeled dilution rate for use against *P. aeruginosa*

Appendix 10 DISPOSAL MEMORANDUM

Disposal Memorandum The following document can be submitted on USDA letterhead if approved landfill operators require some form of documentation.

MEMORANDUM FOR RECORD

March 28, 2003

Based on a review of available data, consultation with the US Environmental Protection Agency (EPA) Office of Solid Waste Management and the APHIS Safety, Health, Environmental and Security Branch, we have determined that *Ralstonia solanacearum* race 3 biovar 2 does not present a risk to human health or the environment, with the exception of certain agricultural crops. We have based our conclusion on the following findings:

- *Ralstonia solanacearum* race 3 biovar 2 is not a human or animal pathogen.
- EPA reports that *Ralstonia solanacearum* race 3 biovar 2 is not considered a hazardous or medical waste.
- EPA reports that it can be disposed of in permitted solid waste.
- It poses no threat to ground water.
- There is a potential risk of infection for certain agricultural crops, especially potatoes but also tomatoes and peppers and other solanaceous plants.
- It can be established in the environment through contamination of natural waterways, soil, and host plants.

In order to prevent the release of *Ralstonia solanacearum* race 3 biovar 2 to the environment and to protect at-risk crops, APHIS requires the following mitigations in its *Ralstonia* eradication action plan:

- Waste (whole plants, plant material, soil and equipment) shall be:
  - double-bagged in securely sealed, leak-proof plastic bags;
  - disposed of in a State, local or tribally permitted solid waste landfill;
  - covered to a depth of two feet with soil at landfill.
- A Federal or State officials shall witness disposal at the landfill to ensure proper handling.

Copies of the action plan are available at the APHIS web site at:
http://www.aphis.usda.gov/ppq/ep/ralstonia/index.html

Appendix 11 Contributors and Consultants

Contributors The following people provided input and/or reviewed these New Pest Response Guidelines.

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