Executive Summary

Updates to program requirements for importing *Ralstonia solanacearum* race 3 biovar 2 host plants under the USDA-APHIS Ralstonia Exclusion Program Framework

February 2024

APHIS is creating a program update for imports regulated for select agent *Ralstonia* solanacearum race 3 biovar 2 (Rs R3bv2). The update pulls several import protocols into a single framework for all Rs R3bv2 host commodities under the Ralstonia Exclusion Program (REP). This summary highlights changes and clarifications in the 2024 REP compared to the 2007 Minimum Sanitation Protocol for Offshore Geranium Cutting Production. In 2005, APHIS developed the Minimum Sanitation protocol in response to the 1999 and 2003 introductions of Rs R3bv2 on imported geranium cuttings. Rs R3bv2 was listed as a select agent in the USDA Agricultural Bioterrorism Protection Act of 2002. The REP may also refer to commodity specific sub-programs as the *Pelargonium* Program or the Tomato Plantlet Program.

Timeline

As *Pelargonium* cuttings are the highest volume commodity under this program, APHIS plans to roll-out the REP in June 2024, before the next *Pelargonium* shipping season. APHIS is issuing this stakeholder announcement to allow Industry, the National Plant Board, and other constituents a 30-day comment period. APHIS will then review the comments and make any final changes as needed with an intention to implement the new framework in June 2024.

Summary of changes or additions:

- Ralstonia Exclusion Program Updates and Clarifications
 - Clarification of the Operational Work Plan requirement to align with the 2005 rule (70 FR 61351). OWPs will have the bilateral agreement signatory page and any information needed in greater detail than the framework for each NPPO.
 - O Combination of other *Ralstonia solanacearum* race 3 biovar 2 host requirements (e.g., tomato plantlets in growing media) under a single framework.
 - Development of an *Ralstonia* Exclusion Program webpage on the APHIS website.
 The site will include the REP framework, APHIS-approved PCR and water testing protocols, and sampling and testing job aids, and will be updated regularly.
 - Clarification of roles and responsibilities of APHIS, NPPOs, the Cooperator, and the Producer.
 - o Explanation of the facility certification process.
 - o Clarification that facilities must keep records for at least three years.

- o Clarification of Administrative requirements of the NPPO and Producers
- o Clarification of APHIS testing methods for destructive sampling.
- Changes to Rs R3bv2 Protocols
 - o Inclusion of sanitation requirements for callused cuttings.
 - o Inclusion of new APHIS-approved water testing protocols.
 - o Inclusion of requirement for a facility pest management plan.
 - o Expansion of Scouting, Testing, and Sampling guidance.
 - o Expansion of training requirement topics.
 - Expansion of processes for detections of *Ralstonia solanacaerum* in an offshore greenhouse, at the PIS, or in the United States.
 - Requirement that facilities must test samples following ISPM31 Methods of sampling consignments. Facilities must sample a minimum sample size for a 95% confidence level at a 5% disease incidence according to lot size based on a hypergeometric distribution.
 - o Requirement that facilities must test their water source and treated recycled water if used, for *Ralstonia solanacearum* race 3 biovar 2 a minimum of every 6 months using an APHIS-approved testing protocol.
 - Addition of ultra filtration as a primary or secondary filtration treatment option for open-sourced water or treated irrigation water.
 - o Addition of benzoic acid as a new APHIS-approved surface disinfectant.
 - Allowance for reuse of plastic potting bags if treated with an approved growing media treatment or surface disinfectant.
 - Requirement that facilities keep records verifying mother stock plants are free from *Ralstonia solanacearum* race 3 biovar 2.
 - Requirement that facilities maintain records of the manufacturer name and the test's limit of detection when using tests other than RPA and PCR to test for Ralstonia solanacearum race 3 biovar 2.

USDA-APHIS Ralstonia Exclusion Program Framework

Effective Date: June 2024



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Ralstonia Exclusion Program Framework Overview

Effective Date of this Framework Document	[Implementation date]	
Countries Involved	1) United States	
	2) NPPO of participating countries where	
	Ralstonia solanacearum race 3 biovar 2	
	(Rs R3bv2) is known to occur	
Commodity(ies)	Pelargonium spp., Solanum spp., and	
	alternative hosts for propagation originating	
	countries where Rs R3bv2 is known to occur	
Type of Program	Audit-based Offshore Program; annual audit	
	(for Production facilities) or tri-annual audit	
	(for Elite facilities) schedule	
Regulated Pests of Concern	Ralstonia solanacearum race 3 biovar 2	
Mitigation Measures	1) Systems approach	
	2) Pest free places of production	
	3) Molecular testing	
Other Special Considerations	7 CFR, Part 331 Possession, Use, and Transfer	
	of Select Agents and Toxins	
Date of Next Framework Review	On or before five years from effective date	
Date of Last Update	2007	

This framework document details the minimum phytosanitary measures required for the USDA Ralstonia Exclusion Program (REP). The REP intends to mitigate risks of importing propagative plant material that can host select agent *Ralstonia solanacearum* race 3 biovar 2 (Rs R3bv2) into the continental United States from countries where Rs R3bv2 is known to occur. The minimum phytosanitary measures include, but are not limited to, protocols for commodity production, packing, safeguarding, diagnostic testing, treatment, export certification, and shipping the United States. Commodities regulated for this select agent are eligible for import when program participants comply with this framework and with an operational work plan (OWP) signed with the National Plant Protection Organization (NPPO) of the country of origin. OWP agreements with each NPPO may describe in greater detail what mitigation measures are needed at specific production sites to meet REP program standards.

The select agent *Ralstonia solanacearum* race 3 biovar 2 (Rs R3bv2) (7 CFR 331.3) is a damaging pathogen of important agricultural commodities such as tomatoes, potatoes, and eggplants. In production greenhouses, spread of this pathogen can occur through transplanting infected plants, pinching buds off plants without sanitizing, using contaminated tools between cuttings, and irrigating with sub-irrigation or ebb-and-flow systems. Growing plants in certified



greenhouses under a systems approach can effectively mitigate most of the pest risk associated with these practices.

In 2005, APHIS amended the regulations by establishing a certification program for articles of *Pelargonium* spp. and *Solanum* spp. imported from countries where the bacterium *Ralstonia solanacearum* race 3 biovar 2 (Rs R3bv2) is known to occur (70 FR 61351). Importation of Rs R3bv2 host commodities are regulated by APHIS to protect American agriculture from the introduction of quarantine plant pathogens (7 CFR 319 Subpart H). APHIS implemented the *Minimum Sanitation Protocol for Offshore Geranium Cutting Production* in response to the February 2003 introduction of *R. solanacearum* R3bv2 via imported geranium cuttings, provisionally adding the certification program to the regulations with an April 2004 interim rule. Last updated in 2007, the program is intended to help prevent introduction of Rs R3bv2 on imports of *Pelargonium* spp. into the United States. In 2016, the United States approved imports of tomato plantlets (*Solanum lycopersicon*) in APHIS approved growing media from Mexico, with minimum phytosanitary measures to prevent introduction of Rs R3bv2 per the 2005 rule. The REP is a framework that combines the minimum sanitation protocols to exclude Rs R3bv2 already implemented for imports of host taxa and lays the groundwork for potential imports of other alternative host species.

APHIS policies for Preclearance and Offshore Programs apply to the REP. Program participants must meet all requirements of this program as verified by the NPPO of the participating facility and APHIS before exporting program material to the United States. Participants must receive written approval from APHIS Preclearance and Offshore Programs for any deviations from the REP and any associated OWP prior to implementation.

Participation in the REP is mandatory to export hosts of select agent *Ralstonia solanacearum* race 3 biovar 2 (Rs R3bv2) to the United States from countries without *Ralstonia solanacearum* pest-free status. To enter the United States, program material must meet the following general criteria:

- Producers must grow host material in an APHIS-approved greenhouse that is recertified yearly by APHIS.
- Producers must grow host material following the requirements of this framework and the bilateral operational workplan agreement between the United States and the NPPO of the exporting country.
- Consignments must meet all permit and import requirements as stated in 7 CFR 319
 Subpart H and described in the Plants for Planting Manual and the APHIS Commodity
 Import Requirements (ACIR) online database.



For additional information, please refer to the Ralstonia Exclusion Program website [NOT CURRENLTY AVAILABLE] on the USDA, APHIS website, review the Frequently Asked Questions, or contact the program manager. Contact details are listed on the APHIS program website.



Definitions, Acronyms, and Web Addresses

Definitions for phytosanitary terms used may be found in ISPM 5, Glossary of Phytosanitary Terms (IPPC, 2015); RSPM 5, NAPPO Glossary of Phytosanitary Terms (Revised) (NAPPO, 2012); RSPM 24, Integrated Pest Risk Management Measures for the Importation of Plants for Planting into NAPPO Member Countries (NAPPO, 2005); and ISO 9000:2015 Quality management systems-Fundamentals and vocabulary.

ACIR: Agricultural Commodity Import Requirements. ACIR provides a single source to search for and retrieve entry requirements for imported commodities. ACIR information includes lists of APHIS-approved facilities and entities, treatment schedules, inspection procedures, and other necessary information to determine admissibility, without the need to access multiple manuals. ACIR is found at the following web address: https://acir.aphis.usda.gov.

Additional Declaration: A statement that provides specific additional information on a consignment in relation to regulated pests or regulated articles. Importing countries require this statement on phytosanitary certificates.

APHIS: U.S. Department of Agriculture, Animal and Plant Health Inspection Service.

APHIS-approved Laboratory: A laboratory whose personnel have been trained by APHIS on molecular detection methods for *Ralstonia solanacearum*. These personnel should be tested on their proficiency on molecular methods by participating in the Proficiency Testing Program. The lab must also have a valid import permit if diagnostic testing occurs in the United States.

Audit: A systematic examination of the organizational structure, infrastructure, procedures, processes, records management, and resources used by the place of production in implementing the Ralstonia Exclusion Program.

Broker: An entity that purchases or takes possession of plants for planting from an approved place of production for the purpose of exporting those plants without further growing beyond maintaining the plants until export.

Callused Cutting: An unrooted cutting with callus tissue (a mass of large, thin-walled, undifferentiated cells that typically form as a precursor to rooting) (see Unrooted Cutting).

Certificate: A document (e.g., official correspondence) by which an APHIS auditor affirms a production site meets Program requirements.



Commercial Shipment: Goods that are imported for resale purposes or for profit (e.g., cuttings to be grown in a nursery for resale as a whole plant): not for personal use.

Commingled: Consignments in which different commodities (or commodity types) have been mixed within individual sampling units (e.g., boxes).

Commodity: A type of plant, plant product, or other article being moved for trade or other purpose.

Confirmed Detection: An Rs R3bv2 detection can be confirmed using an APHIS-approved testing method for Rs R3bv2 following an initial detection for *Ralstonia solanacearum* (see "detection")

Consignment (or Shipment): A quantity of plants, plant products or other articles being moved from one country to another and covered by a single phytosanitary certificate, when required (a consignment may be composed of one or more lots or taxa).

Cooperator: A person or entity responsible for the Cooperative Service Agreement with APHIS.

Corrective Action Request (CAR): A change request form that documents a non-conformance with a product or process in the Ralstonia Exclusion Program and requests the recipient to identify and remove the root cause of the non-conformity. A corrective action is meant to address the cause of a non-conformity and prevent its recurrence.

Country of Origin: The country where the plants, or plants from the plant products were derived or grown — or where the non-plant articles were produced.

Crown: see "Root Crown."

Detection: An initial detection of *Ralstonia solanacearum* or Rs R3bv2 using an APHIS-approved testing method. For all detections (i.e., anything other than a clearly negative result), the facility must send a sample to an NPPO approved laboratory for Rs R3bv2 confirmatory testing using an APHIS-approved method (see "unconfirmed detection")

Eligible Plant: A plant that meets the prerequisite phytosanitary and programmatic conditions to enter the United States.

ELISA: Enzyme-linked immunosorbent assay. ELISA is a biochemical and serological method using enzymatic reactions to detect and quantify the amount of a specific substance, such as viral proteins/particles, is in a solution.



Full Systems Audit: A systematic examination of the organizational structure, procedures, processes, and resources used by the facility in implementing the Ralstonia Exclusion Program. A full systems audit consists of a review of all components of the Minimum Sanitation Protocol for Offshore Geranium Cutting Production.

Greenhouse: The physical location where plants are grown within, under, or sheltered by structures providing a modified growing condition or protection from pests and the outdoor environment. These structures may include greenhouses, hoop houses, screen houses, shade houses, or other structures that are determined by the NPPO of the exporting country to meet the minimum operating requirements of the Ralstonia Exclusion Program.

Hybrid: The offspring of two plants or animals of different species or varieties, such as the Zonal Geranium (*Pelargonium x hortorum*) that comes from a cross of *P. inguinans* and *P. zonale*).

ISPM 31 Methods of Sampling Consignments: Consult ISPM 31 Methods for sampling consignments, Table 1, to determine the number of samples to be collected to have a 95% confidence of detecting 5 percent disease incidence. The table is located at the following web address: https://www.ippc.int/en/publications/83473/.

NAPPRA: Not authorized pending pest risk analysis. A category of regulations governing the importation of plants for planting, commonly known as the Q37 regulations.

National Plant Protection Organization (NPPO): The official service established by a government to discharge the functions specified by the International Plant Protection Convention.

Non-compliance: Activities or products found to be contrary to, or in violation of, APHIS' import regulatory requirements.

Non-conformance: Activities or products found to be contrary to, or in violation of, the Ralstonia Exclusion Program requirements as described in this document.

PCR: Polymerase Chain Reaction. PCR amplifies a specific segment of DNA.

Program: The Ralstonia Exclusion Program (referred to as the Program or REP) is the implementation of the Minimum Sanitation Protocol Offshore Geranium Cutting Production that allows for the export of *Pelargonium* unrooted cuttings or other *Ralstonia* host material to the United States from APHIS-approved Facilities. This document may also refer to commodity-specific sub-programs, such as the Tomato Plantlets in Growing Media Program.



Phytosanitary Certificate: A document, including electronic versions, that is related to a restricted article not more than 15 days prior to shipment of the restricted article from the country in which it was grown and that: (1) Is patterned after the model certificate of the International Plant Protection Convention, a multilateral convention on plant protection under the authority of the Food and Agriculture Organization of the United Nations (FAO); (2) Is issued by an official of a foreign national plant protection organization in one of the five official languages of the FAO; (3) Is addressed to the national plant protection organization of the United States (Animal and Plant Health Inspection Service); (4) Describes the shipment; (5) Certifies the place of origin for all contents of the shipment; (6) Certifies that the shipment has been inspected or tested according to appropriate official procedures and is considered free from quarantine pests of the United States; (7) Contains any additional declarations required in the Plants for Planting Manual; and (8) Certifies that the shipment conforms with the phytosanitary requirements of the United States, and is considered eligible for importation pursuant to the laws and regulations of the United States.

Place of Production (or Facility): Any premises or collection of greenhouses operated as a single production or farming unit. Specific to the Ralstonia Exclusion Program, a place of production or facility is "a contiguous property that is used to produce vegetative cuttings of plants for planting."

Plant Pest: Any living stage of any of the following that can directly or indirectly injure, cause damage to, or cause disease in any plant or plant product: a protozoan, a nonhuman animal, a parasitic plant, a bacterium, a fungus, a virus or viroid, an infectious agent or other pathogen, or any article like or allied with any of these articles.

Plant Unit: The smallest unit in the inspection unit (e.g., cutting, plant, stem).

Plants for Planting: Plants intended to remain planted, be planted, or be replanted. The Plants for Planting Manual is located at the following web address:

https://www.aphis.usda.gov/import_export/plants/manuals/ports/downloads/plants_for_planting.pdf.

PPCDL: Plant Pathogen Confirmatory Diagnostics Laboratory. APHIS's official diagnostics laboratory located in Laurel, Maryland.

Producer: A person or entity responsible for a place of production or production site.



Production Site: A defined portion of a place of production that is managed separately for phytosanitary purposes and utilized to produce a commodity. This may include the entire place of production or portions of it. Examples of portions of places of production are a defined orchard, grove, field, greenhouse, screenhouse, or premises.

qPCR: Quantitative polymerase chain reaction, or quantitative real time PCR.

Quarantine Pest: A plant pest or noxious weed that is of potential economic importance to the United States and not yet present in the United States or present but not widely distributed and being officially controlled.

RPA: Recombinase polymerase amplification.

Regulated Plant Pest Table: The list of U.S. Regulated Plant Pests is found on the APHIS website at https://www.aphis.usda.gov/aphis/ourfocus/planthealth/import-information/rppl/RPPL-Table.

Root Crown: A root crown, also known as the root collar or root neck, is that part of a root system from which a stem arises. The area of the root crown is usually located around or at the soil level and can be vaguely or clearly apparent. *Ralstonia solanacearum* race 3 biovar 2 cells tend to concentrate in the stem near the root crown of infested plants.

Rs R3bv2: *Ralstonia solanacearum* race 3 biovar 2. For more information about Ralstonia, including signs and symptoms, please refer to the APHIS Plant Pest and Disease Programs *Ralstonia* website at: https://www.aphis.usda.gov/aphis/ourfocus/planthealth/plant-pest-and-disease-programs/pests-and-diseases.

Select Agent: A biological agent or toxin that has been determined to have the potential to pose a severe threat to public health and safety, to animal and plant health, or to animal or plant products. Select Agents are regulated by the Federal Select Agent program.

Suspension: An action taken where the facility is not allowed to ship material under the Ralstonia Exclusion Program. A participating Program facility can be suspended if found in violation of any condition of the Program and is unable to implement corrective actions in a timely manner and maintain the required phytosanitary conditions of entry. The facility will be removed from the list of approved Program facilities.



Unrooted Cutting: A section of a plant that is used for propagation without roots. Some unrooted cutting may have callus tissue (a mass of large, thin-walled, undifferentiated cells that typically form as a precursor to rooting, see Callused cuttings). However, for the REP, unrooted cuttings are free of callus tissue, while cuttings with callus are specifically called "callused cuttings".



1 Regulated Articles

- 1.1 Regulated Pests
 - 1.1.1 Select Agent *Ralstonia solanacearum* race 3 biovar 2 (Rs R3bv2) as designated in 7 CFR 331.3.
- 1.2 Regulated Commodities
 - 1.2.1 Host commodities for propagation originating from a country where Rs R3bv2 is known to occur unless they originate from an established Rs R3bv2 pest free area. Host commodities include:
 - 1.2.1.1 *Pelargonium* spp. for propagation
 - 1.2.1.2 *Solanum* spp. for propagation

2 Relevant Authorities and Agreements

- 2.1 Regulations
 - 2.1.1 Phytosanitary conditions for the import of Program commodities from each country are set forth in the United States Code of Federal Regulations (CFR), Title 7: Agriculture, Part 319 Foreign Quarantine Notices, Subpart H Plants for Planting (7 CFR 319.37) and in the Agricultural Commodity Import Requirement (ACIR) Online Database.
 - 2.1.2 Articles accompanied by unmanufactured wood articles, or packaging materials, including wood packaging material, are subject to the International Plant Protection Convention's International Standards for Phytosanitary Measures (ISPM 15) and APHIS' regulations under 7 CFR, Part 319, Subpart I Logs, Lumber, and other Unmanufactured Wood Products, and Subpart N Packaging Materials. These articles may be subject to port of entry compliance verification.
 - 2.1.3 Articles and conveyances are subject to inspection requirements as described in 7 CFR Part 330 Federal Plant Pest Regulations; General; Plant Pests; Soil, Stone, And Quarry Products; Garbage and Part 352 Plant Quarantine Safeguard Regulations.
 - 2.1.4 Articles are subject to port of entry document verification, and may be subject to other monitoring, physical inspection, or other actions at U.S ports of entry as deemed necessary by the Department of Homeland Security, U.S. Customs and Border Protection (CBP), U.S Fish and Wildlife Services, and other pertinent Federal regulatory agencies.



- 2.1.5 Unless otherwise stated, articles must enter through Department of Homeland Security, U.S. Customs and Border Protection (CBP) authorized ports located in proximity to Plant Protection and Quarantine (APHIS) Plant Inspection Stations where articles are subject to document verification and physical inspection per APHIS and CBP policies.
- 2.1.6 Articles suspected or confirmed to have Select Agent *Ralstonia* solanacearum race 3 biovar 2 are subject to the requirements of 7 CFR, Part 331 Possession, Use, and Transfer of Select Agents and Toxins. These and other applicable U.S. Regulations may be accessed at http://www.ecfr.gov.

2.2 Bilateral Agreements

2.2.1 Commodities in the Program are subject to the standards of this framework and with a bilateral operational work plan (OWP) agreed to by the National Plant Protection Organization (NPPO) of the country of origin and the U.S. Department of Agriculture (USDA), Animal Plant Health Inspection Service (APHIS).

3 Participants and Responsibilities

- 3.1 The U.S. Department of Agriculture (USDA), Animal Plant Health Inspection Service (APHIS)
 - 3.1.1 APHIS will sign a bilateral agreement for the Program with the exporting NPPO.
 - 3.1.2 APHIS will provide technical and operational guidance to ensure phytosanitary security and Program integrity.
 - 3.1.3 APHIS will conduct the initial full-systems facility audit and subsequent, mandatory, annual recertification audits of the registered greenhouse production sites with the NPPO.
 - 3.1.4 APHIS will issue an Import Permit and provide guidance to the Department of Homeland Security (DHS), Customs and Border Protection (CBP) to facilitate the authorized movement of the shipments from the port of entry to the importing greenhouse.
 - 3.1.5 APHIS reserves the right to change and amend this Program framework and will communicate changes to participants prior to establishing new requirements.



- 3.1.6 APHIS-Plant Inspection Stations (PISs) will provide final clearance of the consignments at the ports of entry in the United States. Shipments must enter at a port of entry with a PIS.
- 3.1.7 The PIS clearance process may include review and approval of import documents, verification of consignment contents, inspection of conveyance, phytosanitary inspection, and molecular diagnostic testing of program commodities. Consignments will remain on hold while port inspectors complete the clearance process.
- 3.1.8 The PIS is responsible for releasing the consignments into U.S. commerce.
- 3.2 National Plant Protection Organization (NPPO) of the Exporting Country. The NPPO Must:
 - 3.2.1 Sign a bilateral agreement for the Program with APHIS.
 - 3.2.2 Administer the Program and supervise approved facilities and cooperators under this Program.
 - 3.2.3 Review, approve, certify, and register production facilities participating in the Program and assign each facility a registration code or number.
 - 3.2.4 Provide to APHIS the list of registered facilities that may participate in the Program, 30 days prior to the start of each primary export season or when requested after an APHIS Stakeholder audit invitation notice.
 - 3.2.5 Only issue phytosanitary certificates for Program material originating from places of production that appear on the APHIS-Approved Facility list for the Program in the ACIR online database.
 - 3.2.6 Monitor Program activities to ensure conformance to this framework and related bilateral agreements. (NPPO authorized personnel can complete this task.)
 - 3.2.7 Conduct an audit at each participating facility at least once per calendar year to verify conformance to the Program. The NPPO may conduct an audit concurrently with the APHIS recertification audit.
 - 3.2.8 Review, monitor or conduct plant sample and water testing activities of each participating facility a minimum of once per month during the growing and shipping season to verify conformance to the Program. (NPPO authorized personnel may complete this task.)



- 3.2.9 Inspect Rs R3bv2 host commodities in the production greenhouse no more than 30 days prior to export when issuing phytosanitary certificates. The NPPO must only certify plants found free from evidence of quarantine pests and disease.
- 3.2.10 Inspect Rs R3bv2 host commodities scouting and testing records prior to export certification at a minimum of every 30 days during the growing and export season. The NPPO must only certify plants found free from evidence of quarantine pests and disease.
- 3.2.11 Notify APHIS via official communication of any Producer requests for changes or deviation from the protocol before implementation. (Examples include treatments, disinfectants, initiating the use of recycled water).
- 3.2.12 Inform APHIS immediately of any Ralstonia solanacaerum detections.
- 3.2.13 Immediately suspend export certification for Program material from registered places of production or production sites associated with any confirmed Rs R3bv2 detection as outlined in <u>Appendix 4</u>.
- 3.2.14 Retain required records for three years and make them available to APHIS upon request.

3.3 Cooperators

- 3.3.1 Cooperators include a person or entity responsible for the Cooperative Service Agreement with APHIS.
- 3.3.2 Per APHIS' regulations §319.37-22 the cooperator will reimburse APHIS for the cost of conducting Program audits or site visits. The cooperator will sign a Cooperative Service Agreement and APHIS will establish a trust fund account to receive funds.
- 3.3.3 Cooperators must retain records related to the Program for three years and make them available to APHIS and the NPPO upon request.

3.4 Producers

- 3.4.1 Producers include a person or entity responsible for a place of production or production site.
- 3.4.2 Producers must follow the Program requirements of this Program and any bilateral operational work plan associated with the exported host commodity from the exporting country.



- 3.4.3 Producers must cooperate with the NPPO of the exporting country in their administration of the Program.
- 3.4.4 Producers must sign a compliance agreement with the NPPO of the exporting country, which enforces the bilateral agreement between APHIS and the NPPO for the Program.
- 3.4.5 Producers must allow APHIS and the NPPO of the exporting country access to the facilities used for the production, processing, and export of Program material and allow access to all records and documents relating to the operations of the Program.
- 3.4.6 Producers must submit an official request to APHIS through their NPPO to obtain written approval for any deviations from this protocol prior to implementation. Examples may include changes to treatments, disinfectants or initiating the use of recycled water.
- 3.4.7 Producers shall inform the NPPO immediately of any *Ralstonia* solanacaerum detections.
- 3.4.8 Producers will bear the costs of APHIS personnel supplying support and oversight to the Program by contributing their cooperator's trust fund.
- 3.4.9 Producers must retain records related to the Program for three years and make them available to APHIS and the NPPO upon request.

4 Program Requirements

4.1 Import Requirements

- 4.1.1 Imports of REP material must meet all the permit and import requirements stated in 7 CFR 319.37, the Plants for Planting Manual, and the APHIS Commodity Import Requirements (ACIR) online database.
- 4.1.2 Producers must not comingle (i.e. include in the same box or container) REP material with non-REP material.
- 4.1.3 Producers may participate in both the REP and the Offshore Greenhouse Certification Program (OGCP). However, shipments of OGCP eligible plants will become ineligible for the program if mingled with (i.e. included in the same shipment) with REP material.
- 4.1.4 The phytosanitary certificate must follow the requirements stated in Chapter 2 of the Plants for Planting Manual and ACIR; it must include NPPO-assigned facility code and any additional requirements noted in ACIR.



4.2 Registered Places of Production

- 4.2.1 A place of production may be either a production facility or an elite facility and must be approved by the exporting NPPO.
- 4.2.2 A production facility produces Rs R3bv2 host commodities for the purpose of commercial production and export to the United States. APHIS must certify production facilities once a year.
- 4.2.3 An elite facility produces Rs R3bv2 host commodities for export to the United States for non-commercial purposes, such as trials or research. APHIS must certify elite facilities every third year unless there are changes in production and export practices, or non-conformities or non-compliance warrant more frequent audits.
- 4.2.4 An elite facility must request reclassification to a production facility before sending any commercial shipments to the United States.
- 4.3 Requirements for Approved Facilities
 - 4.3.1 Process for APHIS-certification of NPPO-approved facilities
 - 4.3.1.1 APHIS must certify all offshore facilities and publish their facility information on the ACIR online database Approved Facility list before they can export Program material to the United States. Certified facility information must include the facility name, physical location, contact information, and the NPPO-assigned facility code.
 - 4.3.1.2 The NPPO of the exporting country must verify that the place of production meets the requirements of the Program and agree to maintain oversight of the facility under the Program by signing a bilateral agreement with APHIS.
 - 4.3.1.3 A facility must submit an official request to APHIS through their NPPO to participate in the Program and to request certification or recertification.
 - 4.3.1.4 To maintain participation in the Program, APHIS must recertify facilities once a year for production facilities or every three years for elite facilities.
 - 4.3.1.5 Minimum standards for certification and recertification are in outlined in this document and may include additional standards in the NPPO-APHIS bilateral agreement. Refer to <u>Appendix 2</u> for an



- example of an audit checklist used during a certification or recertification audit.
- 4.3.1.6 The facility must be associated with a Cooperator to participate in the Program.
- 4.3.1.7 The facility must work with a Cooperator to reimburse APHIS for the cost of conducting Program audits or site visits. The cooperator will sign a Cooperative Service Agreement with APHIS, and APHIS will establish a Trust Fund account to collect reimbursements.
- 4.3.1.8 APHIS will provide participants with a budget estimate for full reimbursement of costs associated with oversight activities, including the inspector's salary, benefits, and travel.
- 4.3.1.9 will deposit funds for the visits into a trust fund established with APHIS prior to facility audit and certification activities. Funds should be deposited into the trust fund account a minimum of two weeks prior to the auditor's expected travel date.

4.3.2 Initial Certification

- 4.3.2.1 Participating facilities must be in a country where the NPPO of the exporting country has signed a bilateral agreement with APHIS for the Program.
- 4.3.2.2 When requesting participation on the facility's behalf, the NPPO must verify the facility meets the requirements of the Program, provide that information to APHIS, and request APHIS conduct an initial certification audit.
- 4.3.2.3 To verify the facility meets Program requirements, the NPPO of the exporting country must ensure that:
 - 4.3.2.3.1 Producers test host plant nursery stock for *Ralstonia* solanacearum following guidance in Section 6. All test results must be sent to APHIS prior to the initial certification audit.
 - 4.3.2.3.2 *Producers* test water sources at least every 6 months for *Ralstonia solanacearum* prior to the initial certification audit (refer to <u>Appendix 5</u>). All test results must be sent to APHIS prior to the initial certification audit.



- 4.3.2.3.3 *Producers* treat growing media in accordance with Program requirements. Growing media treatment records for the current growing season must be sent to APHIS prior to the initial certification audit.
- 4.3.3 Requirement to use the APHIS's Core Message Set
 - 4.3.3.1 Facilities participating in the program must submit the required import information, producer name and the NPPO-assigned facility code using the APHIS's core message set in all consignments from the certified facility. For questions about the APHIS core message set, please contact ACE.ITDS@usda.gov.

5 Production Requirements in the Country of Origin

- 5.1 Plant Production Process for *Pelargonium* Cuttings
 - 5.1.1 All propagation must begin with plant material that has been tested and found free of Rs R3bv2.
 - 5.1.2 Production practices for hosts of *Ralstonia solanacearum* race 3 biovar 2 other than *Pelargonium* spp. must verify that seed, grafted stock and nucleus, increase and production stock are certified and tested to verify freedom from Rs R3bv2.
- 5.2 Vegetative Cutting Production Process
 - 5.2.1 Relevant Terms
 - 5.2.1.1 **Certified Seed**: Seed certified to be free of Rs R3bv2 by an accredited certification body.
 - 5.2.1.2 **Mother Stock**: The propagative source material used to populate the Nucleus Block. Mother stock comes from pathogen elimination tissue culture or is grown from certified seed to produce pathogen free propagative plant material. Mother stock must be tested and found free from Rs R3bv2 prior to shipment to the facility (refer to 4.1.1 and 6.2.2).
 - 5.2.1.3 **Nucleus Block**: The first generation (G1) refers to the tissue culture or original nuclear plants that have been tested and found free of pathogens of concern. Nucleus stock plant blocks are subject to periodic renewal.



- 5.2.1.4 **Increase Block**: Generation two (G2) or generation three (G3-second increase block) is plant material propagated from G1 or G2, respectively, in separate, defined growing zones or increase blocks.
- 5.2.1.5 **Production Block**: Generation three (G3) or four (G4) plant material is propagated from G2 or G3, respectively, in dedicated structures or production zones that are separate from increase blocks. Production block plant material is destined for delivery to the nursery's customer.
- 5.2.1.6 Callus Production: Unrooted cuttings harvested for callus rooted cuttings are propagated from Generation three (G3) or four (G4) plant material in dedicated structures or production zones.

 Callused cuttings are unrooted cuttings grown for seven to 16 days after harvest to allow scar tissue formation at the stem cutting base to prepare the plant for root development. Callused cuttings exported to the United States must not include roots.
- 5.2.2 Mandatory Production Requirements
 - 5.2.2.1 Plant production must follow a unidirectional flow of plants for planting which starts with nucleus stock, then increase, then production.
 - 5.2.2.2 Producers can use production stock to create mother stock if plants undergo the same pathogen elimination and testing process used to develop mother stock. Facilities must provide documentation confirming the pathogen elimination process and testing process, if using production stock to create mother stock.
 - 5.2.2.3 To safeguard against cross-contamination, Program plant material must be grown in a dedicated growing area with physical barriers from non-Program plant material. Rs R3bv2 plant taxa susceptible to Rs R3bv2 must not be grown in the same greenhouse compartment with any other plant genus that is restricted from importation into the United States.
 - 5.2.2.4 Facilities must restrict access to nucleus, increase blocks and production blocks to personnel following required preventive hygiene measures.
- 5.3 Plant Production Process for Solanum lycopersicon



5.3.1 Process is described in the Operational Work Plan for the Export of Tomato Plantlets (*Solanum lycopersicon*) in APHIS-Approved Growing Media from Mexico.

5.4 Place of Production Infrastructure

- 5.4.1 Producers must grow and pack all host plant material grown for export to the United States in a pest exclusionary structure registered with the NPPO of the exporting country.
- 5.4.2 Producers should observe the environment surrounding the property and mitigate possible sources of Rs B3bv2 where possible. For example, slopes of local terrain that may allow run-off into the pest-exclusionary area or presence of alternative plant hosts for the pathogen.
- 5.4.3 A registered place of production (i.e., facility) is a greenhouse or collection of greenhouses operated as a single production or farming unit. APHIS regulates places of production.
- 5.4.4 A registered production site consists of a specific greenhouse within a place of production. There can be one or more production sites within a place of production.
- 5.4.5 The NPPO will regulate production sites in accordance with this protocol. Programmatic actions by APHIS following non-conformance or non-compliance found at U.S. ports of entry or in the United States associated with a production site will apply to the place of production (i.e., facility).
- 5.4.6 Greenhouse and Product Grading Area Infrastructure
 - 5.4.6.1 Program material production should occur in a commercial-grade greenhouse.
 - 5.4.6.2 Producers must ensure Program material remains contained within a pest exclusionary structure until loaded for export.
 - 5.4.6.3 Required screens and other physical barriers must prevent entry of pests into the structure with a designated screen mesh size of no more than 3mm x 6mm to address regional pests of quarantine concern.
 - 5.4.6.4 Greenhouse construction must prevent seasonal rain or flood waters from entering the greenhouse and from contacting plant production surfaces and plants.



- 5.4.6.5 A buffer zone composed of gravel or crushed rock on top of double layer weed cloth, or of concrete must surround the outer perimeter of the greenhouses; the buffer zone layer should be a minimum of 2 inches deep. This buffer zone must be at least one meter wide, free of plants, grass, weeds, and exposed soil. Surfaces in the buffer zone must have canals or have a grade on all sides so that water drains away from the greenhouse.
- 5.4.6.6 There must be no exposed soil or dirt in greenhouses or grading areas. Facility floors and walkways must be concrete or crushed rock or gravel on top of a double layer weed cloth. Producers must replace gravel/crushed rock that has become contaminated with soil or plant debris and repair or replace weed cloth that has tears, worn spots or edges that expose the ground. Floors must drain properly to prevent puddles of water.
- 5.4.6.7 Equipment surfaces that regularly encounter stock plants (e.g., ends of hoses or watering wands) must be surface disinfected before each use and may not touch the floor (refer to Appendix 3).
- 5.4.6.8 Stock plants must be rooted and grown in APHIS-approved growing media (refer to Table 7-1 in the APHIS Plants for Planting Manual or ACIR).
- 5.4.6.9 The bottom of benches for growing stock plants must be raised at least 46 cm above the floor surface to prevent contamination from splashing water.
- 5.4.6.10 Entry to the greenhouse must be through a vestibule with closing doors to deter the entry of pests. The vestibule must have two separate doors (screens or solid material) that form a distinct enclosed environment separating the outdoors from the inside of the greenhouse. One door to the vestibule must close before the second one is open. The vestibule must be directly next to or provide direct access to the sanitation station an area equipped with wash station(s), foot bath(s), and protective clothing (e.g., aprons, lab coats, gloves).
- 5.4.6.11 Facility personnel must regularly scout the facility itself for exposed soil, containment breeches, and other environmental concerns. Examples include drainage issues, hoses hung under



irrigation gutters or on the floor, holes in screens, and other signs of compromised phytosanitary measures.

- 5.4.7 Packinghouse and Cold Room Infrastructure
 - 5.4.7.1 Packing area must be clean and disinfected before use.
 - 5.4.7.2 Packing and loading must take place inside a pest exclusionary structure during daylight hours.
 - 5.4.7.3 Physical barriers must prevent entry of pests or hitchhiker insects into the packing area. Examples include a double door system, air curtains, screens with a minimum mesh size of 1.2 mm x 1.2 mm or transparent plastic strips that hang vertically on the doors. There must be no gaps between the wall and floor and between the walls and the ceiling.
 - 5.4.7.4 All packing and shipping containers must be free of debris, soil, weeds, and pests.
 - 5.4.7.5 Packing material must meet import requirements under Section 2-23 "Packing and Approved Packing Material" in the APHIS Plants for Planting Manual.
 - 5.4.7.6 If Producers find live plant pests during packing, the packing must cease until a phytosanitary corrective action (e.g., treatment, re-conditioning of plant material) is implemented that guarantees pest control, and that action must be properly documented. Finding of a live plant pest should also trigger an assessment of the pest exclusionary structure for the packing facility and for the production block that sourced the plants to ensure structural integrity has not been compromised. This assessment should be documented.

5.5 Sanitation Standards

5.5.1 All facility personnel must wear protective clothing and follow the sanitation practices described in this Program to mitigate the risk of introducing regulated pests into production sites. The facility must strictly enforce sanitation practices in greenhouses, cold rooms, and grading facilities. Refer to Appendix 3 for a list of APHIS-approved disinfectants for surface, skin, and clothing.



- 5.5.2 The facility must ensure vehicles only enter the facility after their tires are cleaned and disinfected. The facility may choose to prohibit them from entering entirely.
- 5.5.3 No food is permitted in or near the greenhouses, cold rooms, or grading facilities.

5.5.4 Wash Stations

- 5.5.4.1 Facilities must have wash stations in greenhouses for use by each person entering the greenhouse. All personnel must wash exposed body parts that may encounter plant material with soap or disinfectant prior to entering the greenhouse.
- 5.5.4.2 Personnel must disinfect or change any latex or vinyl gloves they use between each use, and between each greenhouse.

5.5.5 Footwear

- 5.5.5.1 Facilities must provide a sanitation area for each person entering the greenhouse to ensure their footwear is clean prior to entering. Facilities must provide footbaths to disinfect footwear as well as tools to brush or rinse footwear free of soil and debris or provide footwear to be worn specifically within production zones. The bottom surface of the footbath shall be rough in texture to facilitate dislodging any debris from the bottom of footwear. This may be accomplished by placement of a mat inside the footbath.
- 5.5.5.2 The volume of disinfectant used in footbaths must be adequate to submerge the soles and lower portions of footwear such that they remain wet for the minimum amount of time identified on the disinfectant manufacturer label to be effective against *Ralstonia solanacearum*. Facilities must change the footbath disinfectant a minimum of twice daily and remove any debris prior to replacement of the disinfectant.
- 5.5.5.3 The facility must maintain a record log at each footbath of disinfectant type and concentration used, and the date, time, and signature of the personnel who were responsible for changing the footbath(s).
- 5.5.5.4 Producers must retain footbath records for three years and make them available to APHIS and the NPPO upon request.



5.5.6 Protective Clothing

- 5.5.6.1 All personnel entering greenhouses must wear protective clothing to prevent employee clothing worn outside the production sites contacting Rs R3bv2 host plants.
- 5.5.6.2 The facility must provide greenhouse-specific sets of protective clothing.
- 5.5.6.3 Personnel must only put on protective clothing after leaving the sanitation station but prior to entering plant production sites.
- 5.5.6.4 If personnel wear protective clothing between greenhouses within a facility, they must cover the protective clothing with an additional layer of protective clothing.
- 5.5.6.5 The facility must store protective clothing in a way that avoids contact with the floor, and ensures the clothing is free of debris, potting media, soil, or plant material.
- 5.5.6.6 The facility must replace or wash protective clothing weekly. The second set of protective clothing must be stored separately from protective clothing worn within greenhouses prior to washing.
- 5.5.6.7 Personnel Hygiene
- 5.5.6.8 All personnel must follow the hygiene and sanitation standards in Section 5.
- 5.5.6.9 While in the greenhouse facility during production, personnel must regularly disinfect their hands and forearms or gloves by dipping or spraying with disinfectant after each definable production unit (e.g., every 10 plants, pots, or cuttings) and between plant varieties.

5.5.7 Tools and Equipment

- 5.5.7.1 Facilities must disinfect carts and other such equipment each day or between activity changes with an APHIS-approved disinfectant (refer to <u>Appendix 3</u>).
- 5.5.7.2 Facilities must disinfect all knives, scalpels, scissors, cutting collection containers, and other equipment that touch plants



- after each definable production unit (e.g., every 10 plants, pots, or 100 cuttings or bag) and between plant varieties and benches.
- 5.5.7.3 Facilities must disinfect hose ends that have been in contact with greenhouse floors or other potentially hazardous surfaces prior to reuse.
- 5.5.7.4 The volume of disinfectant used to surface disinfect tools should be adequate to submerge the entire blade or portions of tools that contact the plants. Disinfect for the minimum time identified on the manufacturer label to be effective against *Ralstonia* solanacearum.

5.5.8 Handling of Cuttings and Traceability

- 5.5.8.1 The facility must demonstrate trace forward and traceback capability to a specified level (i.e., row or bench) at the production site. Waterproof labels must accompany each bag of cuttings with data to trace the cuttings from the production site to the first U.S. customer.
- 5.5.8.2 The facility must harvest cuttings directly into new plastic bags or into plastic containers that can be disinfected between each use. A use is defined as a harvested bag or definable production unit.
- 5.5.8.3 When transferring cuttings to grading or quality control facilities, bags or containers of cuttings must not touch any material that could expose them to U.S. regulated pests and disease.

5.5.9 Greenhouse Floors

- 5.5.9.1 Greenhouse floors and walkways must be cleaned at least weekly to remove debris and weeds. Greenhouse personnel handling stock plants should not retrieve plant parts that fall to the floor (e.g., cuttings, trimmings). Other workers (or same workers after pruning plants or harvesting of cuttings) should remove and dispose of the collected debris daily.
- 5.5.9.2 Facilities must sanitize greenhouse floors at least annually before each new Rs R3bv2 host plant production cycle begins. Floors must drain properly to prevent standing water and meet all other Program requirements.



5.5.9.3 Producers must retain greenhouse cleaning records for three years and make them available to APHIS and the NPPO upon request.

5.5.10 Growing Media

- 5.5.10.1 All Program material must be grown in APHIS-approved growing media (refer to Table 7-1 in the APHIS Plants for Planting Manual).
- 5.5.10.2 Growing media used for Program material must be stored in a manner that prevents contact with the ground, soil, or turf surfaces.
- 5.5.10.3 Growing media may be either new or reused. New and reused growing media must treated before each use and between plantings by any of the following APHIS-approved methods:

5.5.10.3.1 Steam pasteurization

- 5.5.10.3.1.1 Media must reach and uniformly hold a minimum temperature of 80° Celsius for two hours or 120 minutes.
- 5.5.10.3.1.2 A minimum of 10 sensors must be used to monitor temperature reading during treatment.
- 5.5.10.3.1.3 All sensors must reach a minimum of 80° Celsius before the two-hour (120-minute) treatment begins. If any sensor drops below the minimum temperature during treatment, extend the treatment for the equivalent number of minutes or segments when all sensors were not maintaining a minimum of 80° Celsius.
- 5.5.10.3.1.4 Sensors must be placed one foot below the surface. At the bottom, sensors must be kept in four corners at three to -six inches above the bottom.



- 5.5.10.3.1.5 Sensors must be evenly spaced on left, right, and center.
- 5.5.10.3.1.6 Sensors must be tested and calibrated before use.
- 5.5.10.3.1.7 Facilities must keep records of the time, date, temperature, and duration of treatments. Records must show the time the lowest temperature sensor reaches 80° Celsius until 120 minutes has been completed.

 Temperature readings must be recorded at regular intervals between 10 and 30 minutes.
- 5.5.10.4 Producers must retain growing media treatment records for three years and make them available to APHIS and the NPPO upon request.

5.5.10.4.1 Fumigation

- 5.5.10.4.1.1 If methyl bromide is used, the fumigation rate is 3 grams per liter of media for 72 hours at 21° Celsius or above.
- 5.5.10.4.1.2 If metam sodium 3% is used, the concentration must be 50 ml per five-liter pot or bag, kept covered for 72 hours.
- 5.5.10.4.1.3 Facilities must record the time, date, temperature, fumigant concentration, and duration of treatment.
- 5.5.10.4.1.4 Producers must retain fumigation treatment records for three years and make them available to APHIS and the NPPO upon request.

5.5.11 Plant Containers



- 5.5.11.1 Plant containers for all Program material must be disinfected prior to use and between callus batches or plantings.
- 5.5.11.2 Plant containers may be any of the following:
 - 5.5.11.2.1 New plastic bags or pots.
 - 5.5.11.2.2 Recycled plastic pots that have been surface disinfected with an APHIS-approved disinfectant (Appendix 3).
 - 5.5.11.2.3 Recycled plastic bags that have been sterilized along with reused growing media following requirements in section 5.5.11.

5.5.12 Irrigation

- 5.5.12.1 All irrigation water must be free of Rs R3bv2. The water source, storage method, and irrigation system may require water treatment for Rs R3bv2.
 - 5.5.12.1.1 Potable municipal water or water collected from sealed deep wells does not require treatment if the water is used immediately or is stored in tanks that cannot be contaminated by soil or plant material/debris.
 - 5.5.12.1.2 Water from unsealed wells, rainwater collection systems, ponds, lakes, rivers, streams, or any other type of open body of water, as well as recycled or recirculated water, requires treatment for Rs R3bv2 using an APHIS-approved treatment method (refer to 5.5.13.7).
- 5.5.12.2 Facilities must <u>not</u> irrigate plants using ebb and flow, overhead, or flood irrigation systems.
- 5.5.12.3 Irrigation systems must prevent emitters from contacting growing media or must include emitter backflow devices to prevent contamination of the watering system.
- 5.5.12.4 Facilities must hang watering hoses on a hook to prevent contact with the floor, greenhouse benches, or other potential contaminated surfaces. The last four feet or 1.2 meters of watering hoses as well as any other hand-watering tools must be treated with a



- surface disinfectant (<u>refer to Appendix 3</u>) if they touch the floor, plant material, or growing media.
- 5.5.12.5 Facilities must keep records of routine maintenance, any maintenance issues, and breaches that occur in any part of the irrigation system. Records must include the date, exact location, remedial measures taken, and tests performed to ensure the irrigation system remains free of pathogens.
- 5.5.12.6 To treat water, facilities must have a minimum of two independent water treatment systems: a primary system and a secondary back up system.
- 5.5.12.7 Each water treatment system must consist of <u>both</u> a filtration system and a purification system.
 - 5.5.12.7.1 Filtration systems must be either reed-bed filtration, slow sand filtration, or ultra-filtration.
 - 5.5.12.7.2 Purification systems must be one of the following and must be monitored at regular intervals to ensure the system is functioning as required:
 - 5.5.12.7.2.1 Ozonation (0.4 ppm residual ozone for a minimum of four minutes).
 - 5.5.12.7.2.2 Ultraviolet irradiation: 300J/m2 of UV light at 254 nm with at least 50% light transmission.
 - 5.5.12.7.2.3 Peroxygen products: a minimum residual level of four mg per liter of peracetic acid for two minutes by injecting irrigation water during pumping at 15-35 m³ per hour with a commercial formulation of 50-100 ml/m3 of peracetic acid.
 - 5.5.12.7.2.4 Chlorine dioxide: Dosage of 0.1 mg per liter of residual chlorine dioxide sustained for a two-minute minimum reaction time. This may be achieved by injecting irrigation water



- with 5 mg per liter using a chlorine dioxide generator.
- 5.5.12.7.2.5 Ultra filtration: ultra filtration based on particle size.
- 5.5.12.8 Facilities must maintain records verifying purification methods listed in <u>Section 5.5</u> are operating within desired parameters (i.e., chlorine injections levels within required dosage, UV intensity maintained at required level).
- 5.5.12.9 Producers must retain irrigation and water purification maintenance records for three years and make them available to APHIS and the NPPO upon request.

6 Monitoring, Testing, and Sampling

- 6.1 Pest and Pathogen Monitoring
 - 6.1.1 Each facility must have a written pest management plan that covers Program standards as well as the following elements:
 - 6.1.1.1 Identification of a designated facility pest manager.
 - 6.1.1.2 Monthly structural inspection procedures to ensure compliance with minimum facility standards (e.g., integrity of insect screening).
 - 6.1.1.3 Procedures for the inspection for incoming plant material before the plant material enters the production site.
 - 6.1.1.4 Pest monitoring and control program procedures appropriate for the Program commodity and country of origin and to mitigate against the introduction of pathogen-carrying insects, mollusks, pathogens, and any other pest of concern.
 - 6.1.1.5 Procedures for plant scouting, removal, and disposal of infested or infected plants. For known pests and pathogens, schedule scouting according to predicted emergence dates and or key life cycle events. Schedule scouting at intervals that are frequent enough to prevent or manage outbreaks. Indicate percentage of plants inspected per greenhouse. All culled plants should be tested for Rs R3bv2 (refer to Section 4.1).



- 6.1.1.6 Traceability mechanisms to facilitate targeting, port of entry clearance, traceback and trace forward in the event of noncompliance.
- 6.1.1.7 Procedures for the shipping inspection for outgoing plant material.
- 6.1.1.8 Description of the procedures, documentation, and corrective actions if a non-regulated pest or a regulated pest is detected during the inspection of incoming plant material, during plant production, or in the shipping inspection, grading, or packaging of cuttings. Include the procedure to identify and report pests.
- 6.1.1.9 Procedures to control approved personnel and authorized visitor access to the facility. Facilities must maintain lists of approved personnel.
- 6.1.1.10 Procedures for document control. The facility must maintain records of pest monitoring and management activities for three years and make them available to APHIS and the NPPO upon request.

6.2 Plant Testing

- 6.2.1 Ralstonia solanacearum Testing
 - 6.2.1.1 APHIS-approved types of diagnostic testing for *Ralstonia* solanacearum are subject to change based on new information as it becomes available.
 - 6.2.1.2 Personnel conducting the tests should be trained in the method being used and training records should be maintained by the facility.
 - 6.2.1.3 APHIS-approved testing techniques for routine sampling (see 6.2.4) are PCR, qPCR, RPA, ELISA and lateral flow or strip serological test kits designed to detect *R. solanacearum* or Rs R3bv2. Facilities must maintain records of the name of the manufacturer, expiration date, and analytical sensitivity for tests other than PCR, qPCR or RPA.
 - 6.2.1.4 APHIS-approved testing for symptomatic plant samples (see 6.2.3.2) is PCR, qPCR, or RPA.



- 6.2.1.5 The testing procedure, as well as the storage of reagents and kit components, should follow the manufacturer's instructions provided with each test kit. However, tissue samples must be taken from the lowest part of the stem that is above the soil even if the manufacturers' instructions state that leaves are acceptable.
- 6.2.1.6 When plant testing detects *Ralstonia solanacearum*, Rs R3bv2, or is not clearly negative, send the sample to an NPPO-approved laboratory for confirmatory testing.
- 6.2.1.7 The facility must maintain records of disease testing results for three years and make them available to APHIS or the NPPO upon request.
- 6.2.1.8 Testing must be conducted weekly, with samples collected in accordance with ISPM 31 *Methods of sampling consignments* (refer to Table 6-1). All symptomatic culls must be tested in addition to plants selected for weekly sampling. Personnel conducting testing should not include symptomatic culls in the weekly plant sample numbers collected for monitoring.

Table 6-1. Minimum Sample Sizes for Testing by Lot Size

Number of individual plants (lot size)	Number of plants to randomly sample
1,000	57
2,000	58
3,000	58
4,000	58
5,000	59
6,000+	59
200,000+	59

Minimum sample sizes for disease detection at a 95% confidence level in different population sizes (i.e., lot sizes) given a 5% disease incidence for the population. Sampling sizes were calculated using a hypergeometric distribution. *Source*: ISPM No. 31 (2008) Methodologies for sampling of consignments.

6.2.1.9 Minimum sample size numbers are subject to change as new information regarding *Ralstonia* detection become available.



6.2.2 Mother Stock Testing

- 6.2.2.1 All plant material used for Program material production must be found free of *Ralstonia solanacearum* prior to introduction into an APHIS-approved place of production.
- 6.2.2.2 APHIS-approved testing for mother stock plant samples is PCR, qPCR, or RPA.
- 6.2.2.3 Places of production must maintain records verifying mother stock (G1 or earlier) plants are free from Rs R3bv2. Places of production should maintain documentation verifying imported mother stock, grafted stock or seed was tested and found free from Rs R3bv2 prior to shipment to the facility.
- 6.2.3 Scouting and Testing of Symptomatic Plants
 - 6.2.3.1 Places of production must scout plants weekly for signs of wilt.
 - 6.2.3.2 Places of production must test all suspect or symptomatic plants using PCR in addition to the representative sample tested in 4.1.4. Plant samples must be from whole plants at the lowest part of the plant above the soil (also known as the "root crown"). Symptomatic plants should be destructively sampled.
 - 6.2.3.3 All plants that are discarded for any reason (except when all plants in a greenhouse are destroyed at the end of the production season) must be tested for *Ralstonia solanacearum* prior to destruction. All testing results must be shared with the NPPO and made available to APHIS upon request.

6.2.4 Routine Sampling

- 6.2.4.1 Places of production must test a representative sample of plants for *Ralstonia solanacearum* or Rs R3bv2 weekly during the growing season (refer to Table 6-1, section 6.2.1.8).
- 6.2.4.2 APHIS or the NPPO of the exporting country may require NPPO officials to supervise or execute the weekly sampling and testing.
- 6.2.4.3 The weekly representative sample must:



- 6.2.4.3.1 Be a stratified random sample of plants from across all greenhouses in the place of production (see ISPM 32, section 3.1.3.3 *Stratified sampling*). Producers must do this representative sample in addition to any suspect or symptomatic plants found during routine scouting (see 6.1.1.5).
- 6.2.4.3.2 Include sufficient plant samples to provide a 95% confidence in detecting a 5% disease incidence within the production facility. The maximum number of samples that will need to be collected and tested per week for any lot size is 59 plants, per ISPM 31 *Methods for sampling consignments* (see Table 6-1, section 6.2.1.8).
- 6.2.4.3.3 Be composed of plant stem tissue collected from the lowest part of the plant just above the soil line, often referred to as the root crown. Rs R3bv2 infections concentrate in the lowest part of the stem making leaf and partial stem samples ineffective for testing of non-symptomatic plants.

6.3 Water Testing

- 6.3.1 The water source and treated irrigation water must be tested every six months and prior to the beginning of each new plant production cycle using the APHIS-approved testing protocols to ensure freedom from *Ralstonia solanacearum*. (Refer to <u>Appendix 5</u>).
- 6.3.2 Facilities shall inform the NPPO of the exporting country immediately of any *Ralstonia solanacearum* or Rs R3bv2 detection while performing Program activities.

7 Personnel Training Program

7.1 Training Process

- 7.1.1 At least annually, personnel must take training provided by the Producer in proper practices required to prevent Rs R3bv2 and other regulated pests from entering the facility and becoming established.
- 7.1.2 The facility should limit access to production sites to personnel certified by the training.



- 7.1.3 The Producer must designate a training manager who is responsible for training all personnel in the following:
 - 7.1.3.1 Proper greenhouse and packinghouse practices.
 - 7.1.3.2 Recognizing signs and symptoms of Rs R3bv2 infection.
 - 7.1.3.3 Understanding how Rs R3bv2 can spread from insufficient sanitation and production practices inside the facility.
 - 7.1.3.4 Understanding how Rs R3bv2 can spread from plants outside the facility commonly found in workers' yards or environs.
 - 7.1.3.5 Required sanitary and hygiene practices to prevent pest and disease transmission.
 - 7.1.3.6 As applicable, proper sample collection and testing to meet mandatory routine testing requirements.
- 7.1.4 The training manager must keep records of formal training events, training dates, personnel attendance, and a list of personnel certified by the training.

8 Non-compliance and Non-conformance

- 8.1 Non-compliance
 - 8.1.1 Any Rs or Rs R3bv2 detection at a U.S. port of entry from any consignment originating from a participating facility will result in the following immediate actions:
 - 8.1.1.1 APHIS will issue an Emergency Action Notification (EAN).
 - 8.1.1.2 Shipment will be reexported or destroyed.
 - 8.1.1.3 The facility must stop shipment on the export of Rs R3bv2 host plants from the production site (i.e., greenhouse) associated with the detection while awaiting confirmatory test results for Rs R3bv2 from an NPPO-approved laboratory.
 - 8.1.1.4 Facility will be suspended following an Rs R3bv2 confirmation report from an NPPO-approved laboratory.
 - 8.1.1.5 APHIS will inform the NPPO and Producer of the detection, providing barcode information for the affected variety(ies) to enable traceback to the originating greenhouse.



- 8.1.1.6 The NPPO and Producer must investigate the originating greenhouse, including a review of growing practices within the area and increased scouting, sampling, and testing of plant material within the greenhouse's water source.
- 8.1.1.7 APHIS will require trace forward information for the variety(ies) in the composite sample associated with the detection.
- 8.1.1.8 APHIS may increase PIS testing of shipments from the associated facility until the investigation is complete.
- 8.1.1.9 Before reinstatement of a suspended facility, APHIS will communicate with the NPPO of the exporting country and offshore facility to discuss and agree on mitigation actions to reduce the risk of future *Rs* or Rs R3bv2 detections.
- 8.1.2 Any Rs or Rs R3bv2 detection associated with a participating facility found after consignments enter U.S. commerce will result in the following immediate actions:
 - 8.1.2.1 The U.S. Producer notifies the State Department of Agriculture or APHIS they have detected *Rs* or Rs R3bv2.
 - 8.1.2.2 APHIS will follow the New Pest Response Guidelines and notify State counterparts the results of confirmatory testing.
 - 8.1.2.3 APHIS notifies the Offshore Producer by email of an *Rs* or Rs R3bv2 detection in the United States and provides traceback information and any results from confirmatory testing when available.
 - 8.1.2.4 APHIS notifies the NPPO through official communication after determining the place of production and country of origin of associated plants.
 - 8.1.2.5 APHIS will require trace forward information for the variety(ies) associated with the detection.
 - 8.1.2.6 The associated offshore facility will be suspended following receipt of a Rs R3bv2 confirmed detection report from an NPPO-approved lab.
 - 8.1.2.7 The NPPO and Producer will investigate the originating greenhouse, including a review of growing practices within the area



- and increased scouting, sampling, and testing of plant material within and the greenhouse's water source.
- 8.1.2.8 Before reinstatement of a suspended facility, APHIS will communicate with the NPPO of the exporting country and offshore facility to discuss and agree on mitigation actions to reduce the risk of future pest interceptions.

8.2 Non-conformance

- 8.2.1 Offshore facilities that do not conform with the Program's framework may not be certified/re-certified to participate in the Program. Examples of non-conformance include, but are not limited to, using an incorrect growing media treatment method or rate, insufficient sampling and testing Rs R3bv2 host plants or water sources, and inadequate or non-existent buffer zones. All non-conformities must be resolved and verified prior to any certification or recertification visit.
- 8.2.2 If *Ralstonia solanacearum* or Rs R3bv2 is detected at an offshore facility:
 - 8.2.2.1 The facility must stop shipment on the export of Rs R3bv2 host plants from the production site (i.e., greenhouse) associated with the detection while awaiting confirmatory test results for Rs R3bv2 from an NPPO-approved laboratory.
 - 8.2.2.2 Facility will be suspended following an Rs R3bv2 confirmation report from an NPPO-approved laboratory.
 - 8.2.2.3 Before reinstatement, APHIS will communicate with the NPPO of the exporting country and offshore facility to discuss and agree on mitigation actions to reduce the risk of future pest interceptions.
 - 8.2.2.4 A detection in the export season following a recertification will result in immediate suspension from the Program for the rest of the shipping season until the next mandatory annual recertification audit.
 - 8.2.2.5 Two detections within five years of a recertification results in suspension of the Place of Production in the Program until further notice.
 - 8.2.2.6 Refer to Appendix 4 for detailed procedures following an Rs R3bv2 detection in an offshore place of production prior to export.



8.3 Oversight and Suspension

8.3.1 APHIS program Oversight and Suspension reserves the right to suspend the facility from the Program (i.e., stop shipment of all exports to the United States) based on non-compliance or non-conformance failures.



APPENDIX 1: Example of Corrective Action Request (CAR)

FORM	
CAR Number:	
Program:	Facility Name:
Audit Type:	Facility Code/Number:
Audit Date:	Address:
	Country:
Auditor:	Facility Representative:
Name:	Name:
Phone:	Phone:
Email:	Email:
Step 1: DESCRIPTION of NON-CONFORMA	
Non-conformance Type: Critical Major	r 🗆 Minor
Description:	
Reference to Operational Work Plan/Framework	rk:
Date of CAR:	
APHIS Signature:	Date:
NPPO Signature:	Date:
Facility Representative Signature:	Date:



STEP 2: DESCRIPTION OF CORRECTIVE ACTION

Please submit proposal for corrective action within 7 business days of this form.

Immediate Corrective Action Description:
Proposed Completion Date:
Long-Term Corrective Action Description:
Proposed Completion Date:
Facility Representative Signature:
Date Submitted:
NPPO Signature:
Date Reviewed:
APHIS Signature:
Date Received:



STEP 3: VERIFICATION OF CORRECTIVE ACTION

Note: APHIS will not certify/approve the facility until <u>all</u> corrective actions are closed.

Immediate Corrective Action:
Corrective Action Documentation: □ Acceptable □ Not Acceptable □ N/A
Corrective Action is Acceptable: □ Yes □ No
Follow-up Visit Required: Yes No
Comments:
Long-Term Corrective Action:
Corrective Action Documentation: □ Acceptable □ Not Acceptable □ N/A
Corrective Action is Acceptable: □ Yes □ No
Follow-up Visit Required: Yes No
Comments:
CAR Closed:
APHIS Signature:
Date Closed:

APPENDIX 2: Example of Checklist for Certification Audit- Ralstonia Exclusion Program

1. FACILITY INFORMA	TION				
Facility Name				Date of Inspection	
Country					
Name & Address of Cor	npany (as s	shown on Phyto.	sanitary Certificate	e used for CBP Consignme	ent Clearance)
Is facility listed in the o	nline ACIR	list at the ti	ime of this		
audit?					
Finance Officer Email (a	t parent comp	pany headquart	ters)		
Physical Address of Insp	ected Site	9			
GPS Coordinates of Ins	pected Site	9			
Certified Facility Code					
Facility Manager Nan	ne:				
Email:				Phone:	
Trust Fund Manager (at		Name:			
company)	'	varrie.			
Email:				Phone:	
Total Facility Area			Pelargonium	Area	
(all crops)			1 ciargonium	THE	
Copy of facility map w					
number of employees a			Pelargonium	No. of Greenhous	ses
layout of facility/greenl provided?	iouses				
Total No. of Greenhous	les				
(all crops)	,63		No. Pelargor	nium at Facility	
Total No. of Plants at F	acility		G 1	1': 1 · 1 · (QTT	7 110
(all crops) Greenhouses audited today (GH		. #)			
Total No. of employees			Greenhouses	audited during last	-
(peak season)		inspection (GH #)			
U.S. Importer(s)					
Exporting <i>Pelargonium</i>	cuttings to	o the follow	ing countries:		

(
Dates of ship	ning season				
Dates of peal					
		rvest/prepare cuttings for shipment?			
	ON TUE WED				
		ne facility ship material to the US?			
	ON TUE WED	, I			
Where does	the facility source t	heir nuclear stock/seed? (Provide facility name and physical address)			
	•				
Primary mea	ns of shipment	Land Air Sea			
2. PLACE O	F PRODUCTION IN	RASTRUCTURE			
<u> </u>		are and excludes any external source of Ralstonia			
	m from entering the				
		e tires cleaned and disinfected before entry into the			
-	ite OR are excluded	from the production site?			
Disinfected		Excluded			
		thin an enclosed greenhouse during all stages of growth?			
	There is complete separation between greenhouses?				
		a block greenhouse during a single workday?			
_	_	e unit that is used solely for <i>Pelargonium</i> grown in			
	to USDA standard				
3. GREENH	OUSE BUFFER ZON				
Sanitation	Buffer zone free of soil?	of grass, weeds, dicotyledonous plants, and exposed			
Size	There is at least a	one-meter buffer zone around the entire greenhouse?			
	Buffer zone is slo	ped away from the greenhouse or has canals to prevent			
Slope	water from enteri	ng the greenhouse?			
Бюрс	Greenhouse const	ruction is designed to prevent seasonal rain or flood			
	waters from enter	ing the greenhouse?			
Composition	Gravel Crushed	Rock Concrete Weed Cloth			
Composition	Composition Other:				
4. GREENH	OUSE CONSTRUCT	ON			
	Тор	Glass Polycarbonate Polyethylene			
	Тор	Other:			
	Sides	Glass Polycarbonate Polyethylene Screen			
Material		Other:			
	Describe conditi	on or screen size (optional)			

	All vents and openings in greenhouse are covered with screening to					
	prevent the entry of quarantine pests?					
5. ENTRY AND SANITATION STATION						
	Single entrance into the greenhouse?					
	Direct access to vestibule with wash station?					
Entry	Vestibule area with closing double-door system?					
,	Entry to production site restricted to authorized personnel?					
	Personal items stored before entry to greenhouse?					
	There is a footbath with rough bottom surface prior to entering the greenhouse?					
Footbath	Disinfectant/ concentration:					
	Volume covers soles and lower portions of footwear?					
	Disinfectant is changed at least twice daily?					
	If no, explain:					
	Time and date of disinfectant change is recorded/logged?					
	There is a sink or disinfection station for handwashing prior to entering the production site?					
	Disinfectant/ concentration:					
Wash/						
Disinfection	Sink drains immediately to outside of production site?					
Stations	Water source for sink: (Y or N next to selection) Sealed Well Municipal Other:					
	Treatment:					
	Workers wear latex or vinyl gloves?					
Dressing	There is a protective clothing dressing area outside the production site?					
Area	Protective clothing stored to prevent contact with the floor?					
Protective Clothing	Types (circle) Washable lab coats Washable or disposable aprons Washable or disposable aprons over lab coats					
	Other:					

	Protective clothing dedicated to each greenhouse and removed before						
	exiting? If lab coats are worn between greenhouses, are they covered with or exchanged for an apron at each greenhouse? If no, explain: Clothing is maintained free of debris, potting media, soil, or plant material? Protective clothing washed in detergent weekly or replaced weekly in the case of disposable aprons? Washed Replaced Disinfected						
	Other:						
6. GREENHO	OUSE PRODUCTION						
	Floors	Gravel Crushed stone Concrete Other:					
	Walkways	Gravel Crushed stone Concrete Other:					
	Is exposed soil pro	esent on floors or walkways?					
	If yes, describe:						
	Debris-free?	Weed-free?					
		Free of longstanding puddles of water?					
	Y0 1 11						
	If no, describe:						
Floors and Walkways	Elassa and svaller	average along a day and at locat weed laby (i.e. average weed a d					
waikways	Floors and walkways are cleaned at least weekly. (i.e., swept, washed, brushed)						
	Describe:						
	Describe.						
	If floors and walk	ways are cleaned daily, is debris removed after or					
	during harvesting	·					
	Floors and walkways sanitized at least annually? How Often?						
	Disinfectant/						
	concentration:						
	Plants elevated at	least 46cm above greenhouse floor? How high?					
		ne floor come in contact (e.g., splashing, watering) with					
	plants or benches						
Production	All hard surfaces	(e.g., floors and benches) were sanitized prior to use?					
Surfaces	Disinfectant/						
	concentration:						
	Production surface design and composition ensures drainage?						
	1 Toddetion surface design and composition ensures dramage:						

	Can irrigation water make pot to pot contact?					
	Ebb and flow irrigation system present?					
	Evidence of flood or sub-irrigation system?					
	Ç .					
	Emitters are separate from media in pots?					
	Emitters equipped with backflow devices or raised above media?					
	Does the facility use overhead watering? If yes, describe:					
Irrigation						
System	Hose ends (last 1.2 meters) and irrigation nozzles are off greenhouse					
	floor or production surface?					
	If contact is made, hose and watering equipment treated with surface					
	disinfectant?					
	Disinfectant/					
	concentration:					
	Are hands or gloves and forearms disinfected by dipping or spraying with					
	approved disinfectant?					
	approved distinectant:					
	Disinfectant/					
Personal	concentration:					
Hygiene	Hands or gloves are disinfected every tray of <i>Pelargonium</i> cuttings or					
	definable production unit?					
	Production Unit					
	Food excluded from greenhouse?					
	Carts and collection baskets sprayed with disinfectant after each bag?					
	Disinfectant/					
	concentration:					
	Tools for harvesting or processing <i>Pelargonium</i> cuttings (e.g., knives) soaked in disinfectant prior to use?					
	1					
	Tools for harvesting (e.g., knives) soaked in disinfectant definable production unit?					
	Production Unit					
T 1 1						
Tools and Equipment	Tools for harvesting or processing <i>Pelargonium</i> cuttings (e.g., knives)					
Equipment	rotated between each stock plant or stock plant container/tray?					
	If No, answer questions below:					
	Are tools permanently assigned to a specific bench and appropriately rotated and disinfected between plants on that bench?					
	Are tools appropriately rotated and disinfected between plants in a					
	definable production unit? Production Unit					
	What is the total number of knives stored in a container used for					
	harvesting?					

	Disinfectant volume adequate to submerge entire blade and portions of					
	tools that contact plants?					
	Only new plastic bags or disinfected plastic containers are used for					
D /	collection of harv	ested plants	?			
Bags/ Containers	Plastic containers	are disinfec	ted before being reuse	d?		
Containers	Disinfectant/					
	concentration:					
	~ 1		s labeled with non-wat	er-soluble	ink?	
	Labels accompany each bag/container of cuttings?					
	Label system allo	ws trace for	ward through rooting s	tations or	directly	
Cuttings	from farm to first	wholesale c	ustomer?			
Cuttings	Label system allo	w traceback	to the production site	where they	7	
	originated?					
	_		tact soil or other mater		ng	
	Ralstonia when tr	ansferred to	grading facilities or co	old room?		
		control (cir	cle one) is conducted d	luring harv	est at the	
	bench?					
	-	-	ontrol (circle one) cond	lucted in the	ne	
	greenhous	e at an inspe	ection table?			
	b.) Is Grading / Quality control (circle one) conducted in a separate)	
	room?					
Grading/	Grading/Quality control facilities satisfy same conditions as greenhouses wit				ith	
Quality Control/	respect to:					
Packing	Wash Stations		Hand Washing		Footbath	
facility	Protective		Personal Hygiene		Tools	
	Clothing		1 cisonal frygiene			
	Handling of		Floors		Production	
	production stock				Surfaces	
	Water Treatment		Training Personnel			
			e disinfected between b	oags or con	ntainers of	
	cuttings processed					
7. GROWIN	G MEDIA					
	Growing medium is APHIS-approved (e.g., Scoria, Volcanic rock,					
		ing mix, des	scribe composition)?			
	Describe:					
Type						
V I						
	IC 1. '1					
	If no, describe:					
Cofoguardina	Is madia and/ann	ata atamad as	a goil/dirt or turf gurfac	.og?		
Safeguarding	is media and/or po	ois stored of	n soil/dirt or turf surfac	es:		

	Is media sterilized?						
	Method of tre	atment (Enter Yes or No for relevant treatments)					
	Steam: two hours after all ten sensors reach 80°C						
		Fumigation: Methyl Bromide (3g/liter for 72h @ ≥70°F)					
Sterilization	Fumigation: Metam Sodium (e.g., Vapam, 3% (equivalent to 50 ml pe						
		bag covered for 72 hours)	1				
		gated on a non-porous surface?					
		ature, and duration of treatment at all sensors is					
		ged for growing media treatment? (records maintained for					
	three years) Select One:	Plastic note Plastic bacc					
		Plastic pots Plastic bags new each year?					
	If no, describe						
	11 110, uescribe	C OCIOW.					
Containers							
8. WATER T	ı						
	Municipal	Deep, Sealed Well					
	Other:						
	If "other," des	scribe and indicate treatment:					
	If water is sto	red, is storage such that there is no opportunity for water to					
		ted by soil or plant material/debris?					
		a reed-bed filtration and/or slow sand filtration system?					
Source		tem used in combination with one of the following: (Yes or No)				
		Ozonation (0.4 ppm residual ozone for ≥4 minutes.)					
		Ultraviolet irradiation (300J/m² UV @ 254 nm, ≥ 50%					
		transmission)					
		Peroxygen products (residual level ≥ 4mg / liter per aceti					
		2 min.) (Achieved at injection 15-35m ³ /hour with concen	tration of				
		50-100ml/m3 of peracetic acid.)					
		Chlorine dioxide (residual level ≥ 0.1 mg / liter chlorine d					
		for ≥ 2 min.) (Achieved with 5mg/L injection in irrigation	n water				
		with CO ₂ generator.)					
		Ultra Filtration (per particle size.)					

	Independent backup water treatment system in place in case primary system fails?						
	Is there a log of any routine inspection, breaches, or maintenance issues in the irrigation system kept on site?						
	Water source is tested at least twice a year to ensure freedom from Ralstonia solanacearum? Log of testing kept on site?						
	Detailed description of water	er treatment (use ac	lditional pages if	necessary):			
9. PEST SUR	EVEY AND DETECTION						
	What testing methods, if any	, ,	LISA, PCR, app	roved lateral	flow and		
	strip serological test kits, m	icroscopy):					
	Describe:						
Testing							
Methods	Tests conducted at approved	d/certified:					
1/100110415	In-house laboratory:		NPPO				
			laboratory:				
	Third party laboratory:		Other:				
	Tests conducted by, or unde			ection			
	organization of the country of origin or their designee?						
	Records available for inspec						
	Are samples collected at a r		IPPC ISPM 31?	1			
	Describe sampling program						
	Sampling done prior to first	shipment and wee	kly throughout s	hinning			
	Sampling done prior to first shipment and weekly throughout shipping season?						
Sampling	Approved lateral flow and s	trip serological tes	t conducted corr	ectly?			
	Which plant tissue is tested			J			
	1						
	All suspect wilted plants must be						
	in the lower stem. Testing leaves						
	Plants scouted regularly for			•			
	Test performed prior to dest		ea plants (except	wnen all			
	plants destroyed at end of season).						

	Tests conducted by, or performed under the supervision of, plant				
	protection organization of the country of origin or their designee?				
	Records available for inspection? (maintained a minimum of three years) Production stages sampled:				
	Nuclear/Foundatio		Sampling Rate:		
	n stock (G1)		Sampling Rate.		
	Increase block (G2		Sampling Rate:		
	or G3)		Sampling Rate.		
	Production block		Sampling Rate:		
	(G4)		Sampling Rate.		
10. TRAINING	3				
	Number of Employees of				
	There is a training program covering proper greenhouse procedures and				
	how R. solanacearum spreads?				
	Description of training:				
	How often is training provided? (Yes or No)				
Employee	Annual Monthly Quarterly Weekly Bi-weekly				
Training	Describe:				
	A list of trained personnel is maintained? (maintained a minimum of				
		iei is maintained? (maintai	ned a minimum of		
	three years)				
	List the last date of train		utified in dividualar		
		s limited to trained and cer			
4 (1184844	Facility maintains record of training of personnel and certifications? ARY, CORRECTIVE ACTION REQUESTS and FOLLOW-UP				
1. SUMMAR	RY, CORRECTIVE ACTION	REQUESTS and FOLLOW-	UP		

Observations and						
Summary						
(Attach additional pages, if necessary)						
		ficiencies noted, recomplete Corrective A				agreement on
Corrective Actions						
and						
Follow-up						
I have reviewed detected:	ed and agree	d with the above	and will imme	ediately resolve	outstanding is:	sues
APHIS Inspec	etor:	Signature:	Em	ail:	D	ate:
NPPO Inspector:		Signature:	Em	ail:	D	ate:

Facility Representative:	Signature:	Email:	Date:



APPENDIX 3: LIST OF APPROVED DISINFECTANTS

Table A3-1: Examples of APHIS-approved Surface Disinfectants. Any product used for disinfection should be approved by the NPPO, labeled for use by the proper authority in the country where the facility is located, and must be efficacious against regulated pests. (Last Updated August 2022).

Trade Name	EPA Reg. No.	Active Ingredients	Use Sites
ANTHIUM DIOXCIDE 5% AQUEOUS STABILIZED CHLORINE DIOXIDE	9150-2	Chlorine dioxide	Boot wash/shoe wash
BIO-FRESH CD	9804-3-65516	Chlorine	Greenhouses, hard nonporous surfaces ²
COIL & DUCT SPRAY	9804-3-46463	Chlorine	Greenhouses, hard nonporous surfaces
ECOTREAT	9804-3-7909	Chlorine	Greenhouses, hard nonporous surfaces
ENVIROCON	9804-3	Chlorine dioxide	Greenhouses, hard nonporous surfaces
TOTALINE COIL & DUCT SPRAY	9804-3-40536	Chlorine	Greenhouses, hard nonporous surfaces
ProKure V	87508-3-89334	Sodium chlorite	Greenhouses, hard nonporous surfaces
Virkon	39967-137	Potassium peroxy- monosulfate and sodium chloride	Greenhouses, hard nonporous surfaces, vehicles
Zero Tol	70299-1	Hydrogen dioxide	Greenhouse structures, benches, pots, watering systems, evaporative coolers, storage rooms, ventilation equipment, floors, and other equipment
AFCO 4330	4959-16-833	Iodine	Greenhouses, hard nonporous surfaces
BIOSENTRY IODINE DISINFECTANT	65020-4	Nonylphenoxypoly ethoxyethanol	Greenhouses, hard nonporous surfaces

¹ Disclaimer: Mention of companies or commercial products does not imply recommendation or endorsement by the U.S. Department of Agriculture over others not mentioned. USDA neither guarantees nor warrants the standard of any product mentioned. Product names are mentioned solely to report factually on available data and to provide specific information.

² Hard nonporous surface examples: flower buckets, floors, walls, coolers, benches, and counter tops.

Trade Name	EPA Reg. No.	Active Ingredients	Use Sites
CSAN 2339 IDOPHOR SANITIZER	4959-16-67829	Iodine	Greenhouses, hard nonporous surfaces
OAKITE TRISANITE	4959-16-1020	Iodine	Greenhouses, hard nonporous surfaces
SANI DINE	4959-15-64328	Iodine	Greenhouses, hard nonporous surfaces, shoe/boot wash
WESCODYNE	4959-16-1043	Iodine	Greenhouses, hard nonporous surfaces
WEST AGRO ZZZ DISINFECTANT	4959-16	Iodine	Greenhouses, hard nonporous surfaces
ZEP-I-DINE	4959-16-1270	Iodine	Greenhouses, hard nonporous surfaces
Lonza Formulation S-18	6836-77	Quat. Ammonium	Farm, Poultry, Swine, and Mushroom Premise Sanitation Veterinary Practice/Animal Care/Animal Laboratory Disinfection
MAQUAT 128-MT	10324-112	Quat. Ammonium	Outer clothing, field harvesting equipment, walls/floors of coolers, flower buckets, and greenhouse packing areas
MAQUAT 615-HD	10324-72	Quat. Ammonium	Greenhouses, hard nonporous surfaces
MAQUAT 64 MN	10324-113	Quat. ammonium	Florist shops, wholesale florist, shippers, greenhouse packing areas, flower buckets, floors/walls of coolers, benches, and counter tops
Physan 20	55364-5	Quat. Ammonium	Greenhouses, hard surfaces, lawn and turf grass, seedlings, cut flowers, decorative fountains, pools, birdbaths, and plants
Menno-Florades	Not EPA- approved. Must be approved by the NPPO.	Benzoic Acid	Greenhouses, harvesting equipment

Table A3-2 Example of an APHIS-approved Odor Remover. (Last Updated August 2022).

Trade Name	EPA Reg. No.	Active Ingredients	Use Sites
ProKure G	87508-2-89334	Sodium chlorite	Mold and mildew odor remover

- A3.1. List of active ingredients for skin disinfectants recommended by the Centers for Disease Control.
 - Any product used for disinfection should be approved by the NPPO, labeled for A3.1.1. use by the proper authority in the country where the facility is located, and must be efficacious against plant pathogens. (Last updated 2007).
 - A3.1.1.1. Alcohols (>60% ethanol)
 - Chlorhexidine (0.5%-4% depending on preparation) A3.1.1.2.
 - Chloroxylenol (0.3%-3.75%) A3.1.1.3.
 - A3.1.1.4. Iodine and Iodophors (7.5%-10% povidone-iodine)
 - A3.1.1.5. **Quaternary Ammonium Compounds**

Table A3-3 Examples of specific products labeled for use on skin or clothing in the United **States**. Any product used for disinfection should be approved by the NPPO, labeled for use by the proper authority in the country where the facility is located, and must be efficacious against plant pathogens. (Last updated 2007).

Product	Company	
GX-1027 Antimicrobial Soap	Galloway Chemical	
Hibiclens; Hibistat	AstraZeneca	
Canker Guard	Flo Tech. Inc.	
Csan 154 QT Soap	Bell Chem Corporation	
EcoCare 250, EcoCare 260, EcoCare 350,	Ecolab	
EcoCare 360		
AgriCure; Pure & Clean Antibacterial	International Laboratory Technology Corp.	
Handwash with Germsafe		
FS Antimicrobial Hand Cleaner; FS E-2	ZEP Manufacturing Co.	
Sanitizing Hand Soap; Acclaim Antibacterial		
Liquid Hand Soap		
C-Soap	Genesis Technologies	

APPENDIX 4: PROCEDURES TO FOLLOW WHEN RALSTONIA **SOLANACEARUM IS DETECTED IN AN OFFSHORE GREENHOUSE**

APHIS will adhere to the following standard operating procedures when *Ralstonia solanacearum* (Rs) or Ralstonia solanacearum race 3 biovar 2 (Rs R3bv2) is detected at the offshore place of production either from symptomatic plants or during routine testing.

For all detections (i.e., anything other than a clearly negative result), the facility must send a sample to an NPPO-approved laboratory for Rs R3bv2 confirmatory testing using an APHISapproved method.

APHIS reserves the right to modify the protocol and guidelines at any time. APHIS will notify the NPPO of the exporting country and Producer of any changes to the protocol.

A4.1. The Producer:

- A4.1.1. Must notify the NPPO and APHIS of an Rs or Rs R3bv2 detection within three working days. Notice to the NPPO must include records from the growing season from which the infected plants originated.
- A4.1.2. Must stop shipment on the export of Rs R3bv2 host plants from the production site (i.e., greenhouse) while awaiting confirmatory test results for Rs R3bv2 from an NPPO-approved laboratory.
- A4.1.3. Is immediately suspended from the Program until further notice from APHIS following a confirmed Rs R3bv2 detection report from the NPPO-approved laboratory.
- A4.1.4. Must immediately do the following after a confirmed Rs R3bv2 detection:
 - Stop shipping all Program material to the United States until further A4.1.4.1. notice.
 - A4.1.4.2. Contact APHIS by email or phone within 24 hours and provide variety and greenhouse location information for the tested plant material at the place of production (i.e., facility).
 - A4.1.4.3. Notify APHIS of any shipments in transit to the United States.
 - All in-transit shipments will be tested for Rs R3bv2 upon arrival at the PIS. The shipment may be allowed entry if there are no Rs R3bv2 detections.
 - Supply APHIS consignment data for all shipments from the production A4.1.4.4. facility with a confirmed Rs R3bv2 detection to the United States, unless otherwise directed. The data should include:

- A4.1.4.4.1. traceback information and history of plant movements within each place of production and production site;
- A4.1.4.4.2. the shipping date;
- A4.1.4.4.3. consignee name and address;
- A4.1.4.4.4. ultimate consignee name and address if different than consignee address;
- A4.1.4.4.5. number of plants for each plant type;
- A4.1.4.4.6. and shipment identification information (i.e., air waybill, bill of lading).
- A4.1.5. Must not destroy any Rs R3bv2 host plant material at the place of production other than for the use of diagnostic sampling and testing, unless approved by APHIS.
- A4.1.6. Must conduct an internal audit to determine the source and extent of the Rs R3bv2 infection at the place of production if cuttings have been exported to the United States during the growing season.
- A4.1.7. Must deliver the audit findings report to the NPPO. The audit findings report shall include:
 - A4.1.7.1. Source or cause of infection.
 - A4.1.7.2. Extent of infection through traceback investigation at the place of production.
 - A4.1.7.3. Deviations from established Program requirements or other gaps identified, which may have contributed to the detection of Rs R3bv2.
 - A4.1.7.4. Corrective actions identified by the Producer as part of the internal audit and planned short-term or long-term solutions.
- A4.1.8. Must allow APHIS to participate in the audit and inspections as part of the investigation and recertification of the facility, as needed. The place of production will fund APHIS' audit through their Cooperative Service Agreement and Trust Fund.

A4.2. The NPPO:

- A4.2.1. Must stop shipment on the export of all Rs R3bv2 host plant consignments from the place of production while awaiting the confirmatory results for Rs R3bv2 from an NPPO-approved laboratory.
- A4.2.2. Must notify APHIS of a confirmed Rs R3bv2 detection report from the NPPO-approved laboratory, including the associated facility and varieties:
 - A4.2.2.1. immediately if there are shipments in transit from the associated facility, or
 - A4.2.2.2. within three working days if there are no shipments in transit.

- A4.2.3. Must issue an immediate stop on the export of all Rs R3bv2 host plant consignments from the place of production upon receipt of a confirmed detection of Rs R3bv2 from the NPPO-approved laboratory.
- A4.2.4. Must collect and submit additional samples, in coordination with the producer, to APHIS or other approved NPPO laboratory for Rs R3bv2 confirmatory testing.
- A4.2.5. Must evaluate the results of the Producer's internal traceback investigation.
- A4.2.6. Must conduct independent traceback investigation to identify source and extent of infection and provide results to APHIS.
- A4.2.7. May collect independent samples for testing for confirmatory results during the investigation.
- A4.2.8. May monitor the destruction of infected plants per APHIS' request.
- A4.2.9. Must provide APHIS with facility records from the growing season from which the infected plants originated.

A4.3. APHIS:

- A4.3.1. Will place a "stop shipment" order on the export of Rs R3bv2 host plants from the production site (i.e., greenhouse) while awaiting confirmatory test results for Rs R3bv2 from an NPPO-approved laboratory.
- A4.3.2. May quarantine the entire place of production if not confident that the infestation is confined to the production site.
- A4.3.3. Will suspend the place of production and remove them from the list of approved facilities in ACIR upon receipt a confirmed Rs R3bv2 detection report from the NPPO-approved laboratory.
- A4.3.4. Will request the NPPO of the exporting country hold destruction of plants at the associated place of production until APHIS can review the results of the NPPO and Producer internal investigation.
- A4.3.5. Will conduct trace forwards on all consignments that entered the United States from the associated place of production during the relevant growing season based on consignment data provided by the Producer.
- A4.3.6. Must audit the place of production to remove the facility suspension and add them back to the list of approved facility. APHIS may also audit the place of production during the investigation. The place of production will fund APHIS' audit through Cooperative Service Agreement and Trust Funds.
- A4.3.7. Will review the investigative reports provided by the NPPO of the exporting country and will use the report in conjunction with the recertification audit to determine its recertification decision.
- A4.3.8. Will formally notify the NPPO when the place of production may resume export of Rs R3bv2 host material to the United States.
- A4.3.9. Will enforce consequences of additional Rs R3bv2 detections:

- A4.3.9.1. Additional Rs detections at originating from the place of production may result in another investigation with more stringent scouting, sampling, and testing requirements.
- A4.3.9.2. A confirmed Rs R3bv2 detection in subsequent shipments or in the place of production will result in immediate suspension from the REP for the rest of the shipping season and until recertification. (Refer to Section 8 for additional guidance.)
- A4.3.9.3. Two confirmed Rs R3bv2 detections in shipments or in the place of production within five years will result in suspension of the Place of Production until further notice.

APPENDIX 5: TESTING METHODS FOR THE DETECTION OF RALSTONIA SOLANACEARUM IN IRRIGATION WATER AT OFFSHORE RS R3bv2 Host Plant Greenhouse Facilities

Participating facilities must test their water source, treated irrigation water, and recycled water after treatment for *Ralstonia solanacearum* at a minimum of every six months. Water samples must be sent to an NPPO-approved laboratory. Facilities must use one of the following APHIS-approved methods below to test for *Ralstonia solanacearum* in water:

- A culture-based approach combined with molecular identification, or
- A filtration-based method coupled with molecular detection.

Testing irrigation runoff water for *Ralstonia solanacearum* is not required. If facilities wish to test runoff water for the presence of *Ralstonia solanacearum* in their production stock, APHIS recommends testing via the following method: heterotrophic bacteria (heterotrophic plate count,* see A5.2).

- A5.1. Detection Methods for *Ralstonia solanacearum* in Treated Irrigation Water
 - A5.1.1. For the collection of treated irrigation water to detect Ralstonia solanacearum, water in holding tanks must be collected using sterile tubes or bottles.
 - A5.1.2. The volume of water collected is dependent on the downstream testing method to be used (refer to below).
 - A5.1.3. Duplicate samples must be taken. Samples should be transported in cool, dark conditions (4–10°C) and tested within 24 hours.
 - A5.1.4. The facilities may choose one of the following methods to test for Ralstonia solanacearum in water:
 - A5.1.4.1. A culture-based approach combined with molecular identification tests (EPPO Bulletin 48, 2018)
 - A5.1.4.1.1. Facilities may follow the EPPO Bulletin 48 Standard for testing water. Concentrate collected water samples using one of the following methods:
 - A5.1.4.1.1.1. Centrifugation of 30–50 mL sub-samples and resuspension of the pellet.
 - A5.1.4.1.1.2. Membrane filtration (1 L through a maximum pore size of $0.45 \mu m$) followed by washing.
 - A5.1.4.1.1.3. Perform dilution plating of the concentrated samples on a semi-selective SMSA/Sequeira medium. Test for typical colonies using real-time PCR or conventional PCR (as specified in the bulletin).

- A5.1.4.2. A filtration-based method coupled with molecular detection (PPCDL, 2021)
 - A5.1.4.2.1. Facilities may follow the APHIS PPCDL protocol for testing water. The Work Instruction for this protocol is found on the APHIS program website. The protocol may be updated at any time. Facilities are responsible for using the most updated version of the water testing protocol found on the APHIS website.
 - A5.1.4.2.2. This method uses a filter funnel system to concentrate Ralstonia solanacearum from a 275ml water sample and is coupled with a molecular-based detection (real-time PCR assay). The limit of detection of the entire protocol is 1×101 CFU/ml.
- A5.2. Recommendation for Testing *Ralstonia solanacearum* in Run-off Water for Facilities that Recycle Water
 - A5.2.1. Facilities may choose to recycle irrigated runoff water in efforts to reduce resources and fertilizer entering the local environment.
 - A5.2.2. Run-off water treatment efficacy testing is an effective alternative to testing specifically for *Ralstonia solanacearum*. However:
 - A5.2.2.1. This recommendation does not apply if there are reasons to suspect that there is a source of contamination at the greenhouse level. Facilities must report Rs R3bv2 detections to the NPPO.
 - A5.2.2.2. While recommended for monitoring purposes, runoff water treatment testing is not included in the Program requirements for biannual water testing.
 - A5.2.3. To test run-off water treatment efficacy, APHIS recommends testing the treated water for heterotrophic bacteria (heterotrophic plate count*). The reduction in heterotrophic bacteria is an indirect indicator of pathogen removal (if the pathogen is present) for effectiveness of the water treatment process.
 - A5.2.4. The possible presence of inhibitory substances, such as chlorine, must be considered when testing the treated water. Chemicals such as chlorine may affect heterotrophic plate counts and lead to inaccurate results. Inhibitory substances must be neutralized prior to testing.

Note: Treated run-off water that will be used for irrigation must be tested as described in <u>Section</u> 6.3.

*3M Petrifilm is an available product for this test, but APHIS does not endorse this or other brands.