USDA-APHIS Ralstonia Exclusion Program Framework

Effective Date: June 2024



Table of Contents

Та	Table of Contents i				
Ra	Ralstonia Exclusion Program Framework Overviewii				
De	Definitions, Acronyms, and Web Addresses iv				
1	Regulated Articles1				
2	Relevant Authorities and Agreements1				
3	Participants and Responsibilities2				
4	Ralstonia Exclusion Program Requirements				
5	Production Requirements in the Country of Origin				
6	Monitoring, Testing, and Sampling				
7	Personnel Training Program				
8	Non-compliance and Non-conformance				
AF	PPENDIX 1:Example of Corrective Action Request (CAR) Form				
APPENDIX 2: Example of Checklist for Certification Audit- Ralstonia Exclusion Program32					
APPENDIX 3:List of Approved Disinfectants43					
Ał	APPENDIX 4:Procedures to Follow when <i>Ralstonia solanacearum</i> is Detected in an Offshore place of production				
AF	APPENDIX 5: Testing Methods for the Detection of <i>Ralstonia solanacearum</i> in Irrigation Water at Offshore Rs R3bv2 Host Plant Places of Production				

Effective Date of this Framework Document	[June 1, 2024]
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. and their hybrids, Solanum
	spp., 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

Ralstonia Exclusion Program Framework Overview

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 REP 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 standards.

The select agent Rs R3bv2 (7 CFR 331.3) is a damaging pathogen of important agricultural commodities such as tomatoes, potatoes, and eggplants. In production sites like 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 places of production 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. (including hybrids) imported from countries where the bacterium Rs R3bv2 is known to occur (<u>70 FR 61351</u>). Importation of Rs R3bv2 host commodities are regulated by APHIS to protect American agriculture and natural resources 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 1999 and 2003 introductions of Rs R3bv2 via imported geranium cuttings, provisionally adding the certification program to the regulations with an April 2004 interim rule. In 2016, the United States approved imports of tomato plantlets (*Solanum lycopersicon*) in APHIS-approved growing media with minimum phytosanitary measures to prevent introduction of Rs R3bv2 per the 2005 rule. The REP framework provides a unified standard to address Rs R3bv2 for all propagative hosts.

REP participants must meet all requirements of this REP as verified by the NPPO of the participating place of production and APHIS before exporting REP 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 Rs R3bv2 to the United States from countries without *Ralstonia solanacearum* pest-free status. To enter the United States, REP material must meet the following criteria:

- Producers must grow host material in an APHIS-approved place of production 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 <u>Ralstonia Exclusion Program website</u> on the USDA, APHIS website, review the Frequently Asked Questions, or contact the program manager. Contact details are listed on the <u>APHIS REP website</u>.



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 places of production (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.

NPPO-approved Laboratory: A laboratory approved by a recognized NPPO to test *Ralstonia solanacearum* using APHIS-approved methods. 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 or importing 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). Callused cuttings are unrooted cuttings grown for seven to 16 days after harvest to allow scar tissue formation at the stem cutting base.

Certification: When APHIS affirms a place of production meets REP requirements by issuing an official document (e.g., certification letter or certificate).



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 APHISapproved testing method. For all detections and unexpected results, the place of production 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.



Elite Stock: Advanced breeder or cultivar stock plants that are also typically pathogen free. Elite stock may include patented varieties (see stock plant).

EPPO Bulletin 48: European and Mediterranean Plant Protection Organization (EPPO) Standard "PM 7/21 (2) *Ralstonia solanacearum*, *R. pseudosolanacearum* and *R. syzygii (Ralstonia solanacearum species* complex)," *EPPO Bulletin* Volume 48, Issue 1 (2018): 32-63, https://doi.org/10.1111/epp.12454. This Standard describes a diagnostic protocol for *Ralstonia solanacearum*, *Ralstonia pseudosolanacearum* and *Ralstonia syzygii*, i.e. phylotype/sequevar strain in the *Ralstonia solanacearum* Species Complex (RSSC).

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 APHIS and the NPPO of the exporting country to meet the minimum operating requirements of the Ralstonia Exclusion Program and associated OWP. For this framework, a greenhouse is equivalent to a production site.

Hybrid: The offspring of two different plant taxa bred to produce a new taxon. For example, *Pelargonium x hortorum* is a complex hybrid involving direct crosses and back crosses to multiple *Pelargonium* species. All hybrids involving taxa regulated for Rs R3bv2 are considered subject to the Ralstonia Exclusion Program.

ISPM 31 Sampling methods: International Plant Protection Convention (IPPC), International Standards for Phytosanitary Measures (ISPM) "No. 31 *Methodologies for Sampling of Consignments*," (2008), <u>https://www.ippc.int/en/publications/83473/</u>.

Mingle: Consignments in which different commodities (or commodity types) have been mixed together within the consignment but not within the sampling units (e.g., baggies, boxes, bunches, bundles, etc.)

Mother stock: group of plants from which plant parts are taken to produce new plants (see stock plant).

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.

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: Any premises or collection of production sites (i.e., greenhouses) operated as a single production or farming unit. Specific to the Ralstonia Exclusion Program, a place of production is equivalent to a facility in that it 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/sites/default/files/plants_for_planting.pdf and ACIR

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 area: an area in a place of production that is considered clean for phytosanitary purposes. This may include the entire place of production or portions of it.



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.

REP: The Ralstonia Exclusion Program is a framework that outlines the minimum sanitation protocols to exclude Rs R3bv2 from imports of host taxa to the United States from APHIS-approved Facilities (Places of Production). This document may also refer to commodity-specific sub-programs, such as the Tomato Plantlets in Growing Media Program. The program webpage is located at <u>https://www.aphis.usda.gov/plant-imports/ralstonia-exclusion-program</u>.

RPA: Recombinase polymerase amplification.

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

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: <u>https://www.aphis.usda.gov/aphis/ourfocus/planthealth/plant-pest-and-disease-programs/pests-and-diseases</u>.

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.

Stock Plant: A plant that provides starting material for vegetative propagation. Clean stock plants may come from pathogen elimination tissue culture or be grown from certified seed to produce pathogen free propagative plant material. Sometimes called "mother stock."



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

Unconfirmed detection: An Rs R3bv2 detection that has not yet been confirmed using an APHIS-approved testing method for Rs R3bv2 following an initial detection for *Ralstonia solanacearum* (see "detection").

Unexpected result: results inconsistent with or significantly outside the expected reference range(s). Any test results that do not conform to the expected negative results must be sent to a NPPO-approved lab for Rs R3bv2 testing.

Unrooted Cutting: A section of a plant that is used for propagation without roots. Some unrooted cuttings 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. and hybrids for propagation
1.2.1.2	Solanum spp. and hybrids for propagation

2 Relevant Authorities and Agreements

2.1 Regulations

2.1.1 Phytosanitary conditions for the import of REP 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 <u>http://www.ecfr.gov</u>.

2.2 Bilateral Agreements

2.2.1 The ability to export REP commodities to the United States under this framework is subject to a bilateral agreement between the Animal and Plant Health Inspection Service (APHIS) and the National Plant Protection Organization (NPPO) of the country of origin.

3 Participants and Responsibilities

3.1 The U.S. Department of Agriculture (USDA), Animal and Plant Health Inspection Service (APHIS)

3.1.1 APHIS will sign a bilateral agreement for the REP with the exporting NPPO.

3.1.2 APHIS will provide technical and operational guidance to ensure phytosanitary security and REP integrity.

3.1.3 APHIS will conduct the initial full audit and subsequent, mandatory, annual recertification audits of the registered place of production 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 REP 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 REP 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 REP with APHIS.

3.2.2 Administer the REP and supervise approved places of production, production sites, and cooperators under this REP.

3.2.3 Review, approve, certify, and register places of production participating in the REP and assign each place of production a registration code or number.

3.2.4 Provide to APHIS the list of registered places of production that may participate in the REP, 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 REP material originating from places of production that appear on the APHIS-Approved Facility list for the REP in the ACIR online database.

3.2.6 Monitor REP activities in each facility to ensure conformance to this framework and related bilateral agreements.

3.2.7 Conduct an audit at each participating place of production at least once per calendar year to verify conformance to the REP.

3.2.8 Review, monitor, or conduct plant and water testing activities of each participating place of production a minimum of once per month during the growing and shipping season to verify conformance to the REP.

3.2.9 Inspect Rs R3bv2 host commodities in the production site 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 deviations from this framework or the associated OWP 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 REP material from registered places of production or production sites associated with any confirmed Rs R3bv2 detection as outlined in APPENDIX 4.

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 REP audits or site visits through a trust fund. 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 REP 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 cooperate with the NPPO of the exporting country and the NPPO's administration of the REP.

3.4.3 Producers must follow the requirements of this framework and the associated bilateral agreement APHIS has with the exporting country.

3.4.4 Producers should only work with Brokers that follow the requirements of this REP and any associated bilateral agreements with the exporting country to avoid potential delays or rejection of shipments at the U.S. port of entry.

3.4.5 Producers must sign a compliance agreement with the NPPO of the exporting country, which enforces the bilateral agreement between APHIS and the NPPO and this framework.

3.4.6 Producers must allow APHIS and the NPPO of the exporting country access to the facilities used for the production, processing, and export of REP



material and allow access to all records and documents relating to the operations of the REP.

3.4.7 Producers must submit an official request to APHIS through their NPPO to obtain written approval for any deviations from this framework or the associated OWP prior to implementation. Examples may include changes to treatments, disinfectants or initiating the use of recycled water.

3.4.8 Producers shall inform the NPPO immediately of any *Ralstonia solanacaerum* detections.

3.4.9 Producers will bear the costs of APHIS personnel supplying support and oversight to the REP by contributing to their cooperator's trust fund.

3.4.10 Producers must retain records related to the REP for three years and make them available to APHIS and the NPPO upon request.

4 Ralstonia Exclusion 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 bag) REP material with non-REP material. REP material may be mingled (i.e. in the same consignment) but not comingled.

4.1.3 Producers may participate in both the REP and the Offshore Greenhouse Certification Program (OGCP). However, consignments of OGCP eligible plants will become ineligible for the program if mingled with (i.e. included in the same consignment) 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 place of production code and any additional requirements noted in ACIR.

4.2 Registered Places of Production

4.2.1 A place of production must be registered and approved by the exporting NPPO.

4.2.2 APHIS must certify places of production once a year unless they are classified by APHIS as an elite facility.



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 non-conformities or non-compliances that warrant more frequent audits.

4.2.4 An elite facility will lose its elite classification if there are changes in production and export practices that result in export of commercial shipments to the United States. The place of production must notify the NPPO and APHIS of these changes.

- 4.3 Requirements for Approved Facilities
 - 4.3.1 Process for APHIS-certification of NPPO-approved places of production
 - 4.3.1.1 The NPPO of the exporting country must verify that the place of production meets the requirements of the REP and agree to maintain oversight of the place of production under the REP by signing a bilateral agreement with APHIS.
 - 4.3.1.2 A place of production must submit an official request to APHIS through their NPPO to participate in the REP and to request certification or recertification.
 - 4.3.1.3 APHIS must certify all offshore places of production before they can export REP material to the United States.
 - 4.3.1.4 APHIS must publish place of production information on the ACIR approved facility list before they can import REP material to the United States. The information must include the place of production name, physical location, contact information, and the NPPO-assigned place of production code.
 - 4.3.1.5 To maintain participation in the REP, APHIS must recertify places of production once a year for production facilities or every three years for elite facilities.
 - 4.3.1.6 Minimum standards for certification and recertification are outlined in this document and may include additional standards in the NPPO-APHIS bilateral agreement. Refer to APPENDIX 2 for an example of an audit checklist used during a certification or recertification audit.
 - 4.3.1.7 The place of production must be associated with a Cooperator to participate in the REP.

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- 4.3.1.8 The place of production must work with a Cooperator to reimburse APHIS for the cost of conducting REP 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.
- 4.3.1.9 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.
- 4.3.1.10 Cooperator will deposit funds for the visits into a trust fund established with APHIS prior to any place of production audit and certification activities. Funds should be deposited into the trust fund account prior to the audit.
- 4.3.2 Initial Certification
 - 4.3.2.1 Participating places of production must be in a country where the NPPO of the exporting country has signed a bilateral agreement with APHIS for the REP.
 - 4.3.2.2 When requesting participation on the place of production's behalf, the NPPO must verify the place of production and production site(s) meets the requirements of the REP, provide that information to APHIS, and request APHIS conduct an initial certification audit.
 - 4.3.2.3 To verify the place of production meets REP requirements, the NPPO of the exporting country must ensure that:
 - 4.3.2.3.1 Producers test host plants 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 for *Ralstonia* solanacearum prior to the initial certification audit (refer to APPENDIX 5). All test results must be sent to APHIS prior to the initial certification audit. Certified facilities test water every six months.
 - 4.3.2.3.3 Producers treat growing media in accordance with REP 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 Places of production participating in the REP must submit the required import information, producer name and the NPPO-assigned place of production code using the APHIS's core message set in all consignments from the certified place of production. 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 Rs R3bv2 host plants

5.1.1 All propagation must begin with plant stock or certified seed that has been tested and found free of Rs R3bv2.

5.1.2 All hosts of *Ralstonia solanacearum* race 3 biovar 2 must be regularly sampled and tested to verify freedom from Rs R3bv2.

5.1.3 Plants from any generation of propagation may be exported to the United States if the exported plants are grown in production sites that are inspected and adhere to the requirements of this REP and associated OWP. Defined growing zones used for export must be clearly indicated.

5.2 Relevant Terms

5.2.1 **Certified Seed**: Seed certified to be free of Rs R3bv2 by an accredited certification body.

5.2.2 Elite Stock: The propagative source material used to populate the Nucleus Block. Elite stock comes from pathogen elimination tissue culture or is grown from certified seed to produce pathogen free propagative plant material. Elite stock must be tested and found free from Rs R3bv2 prior to introduction into an APHIS-approved place of production. (refer to 4.1.1 and 6.2.1.1).

5.2.3 **Nucleus Block**: Defined growing zone with elite stock that have been tested and found free of pathogens of concern. Nucleus stock plant blocks are subject to periodic renewal.

5.2.4 **Increase Block**: Defined growing zone with plant material propagated from the nuclear block to increase plant unit volume and not intended for export.

5.2.5 **Production Block**: Dedicated structures or production zones that are separate from increase blocks where plant material is propagated and destined for export.



5.2.6 **Callus Production Block**: Dedicated production blocks that are separate from other production blocks and only produce callused cuttings for export.

5.3 Mandatory Production Requirements

5.3.1 Plant production must follow a unidirectional flow of plants for planting which starts with nucleus stock, then increase, then production. Places of production should maintain a production flow diagram that is available to APHIS and the NPPO upon request.

5.3.2 Producers can use production stock to create elite stock if plants undergo the same pathogen elimination and testing process used to develop elite stock. Places of production must provide documentation confirming the pathogen elimination process and testing process, if using production stock to create elite stock.

5.3.3 To safeguard against cross-contamination, REP plant material must be grown in a dedicated growing area with physical barriers from non-REP 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.

5.3.4 Places of production must restrict access to nucleus, increase, and production blocks to personnel certified in required preventive hygiene measures (see 7.1.2).

5.4 Plant Production Process for plantlets

5.4.1 Only *Solanum lycopersicon* plantlets are approved for import at this time. The production process is described in the Operational Work Plan for the Export of Tomato Plantlets (*Solanum lycopersicon*) in APHIS-Approved Growing Media from Mexico.

5.5 Place of Production Infrastructure

5.5.1 Producers must grow and pack all host plant material grown for export to the United States in a place of production registered with the NPPO of the exporting country.

5.5.2 Producers should observe the environment surrounding the property and mitigate possible sources of Rs B3bv2 where possible. For example, divert runoff from slopes away from the pest-exclusionary area or remove alternative plant hosts for the pathogen.



5.5.3 A registered place of production (i.e., facility) is a production site (i.e., greenhouse) or collection of production sites (i.e., greenhouses) operated as a single production or farming unit. APHIS regulates places of production.

5.5.4 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.

5.5.5 Producers must maintain a map of the place of production registered in the REP, including designated production sites and the clean production area(s). This map must be available to the NPPO and APHIS upon request.

5.5.6 The NPPO will regulate production sites in accordance with this protocol. Programmatic actions by APHIS following non-conformance or noncompliance 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).

- 5.5.7 Greenhouse, Product Grading, and Quality Assessment Area Infrastructure
 - 5.5.7.1 REP material production should occur in a commercial greenhouse.
 - 5.5.7.2 Producers must ensure REP material remains contained within a pest exclusionary structure or safeguarded until loaded for export.
 - 5.5.7.3 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, or as determined in the OWP to address regional pests of quarantine concern.
 - 5.5.7.4 Production site construction must prevent seasonal rain or flood waters from entering the production site and from contacting plant production surfaces and plants.
 - 5.5.7.5 A buffer zone must surround the outer perimeter of the production site. A buffer zone must be an area at least one meter wide that is free of all 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 production site. Acceptable buffer zone materials include a 2-inch deep layer of crushed rock or gravel, a layer weed cloth, concrete, or a combination thereof.
 - 5.5.7.6 Production site floors and grading areas must be free of all plants, grass, weeds, and exposed soil. Acceptable materials for floors



and walkways include concrete, crushed rock or gravel, a layer weed cloth, or a combination thereof. Producers must replace crushed rock or gravel that has become contaminated with soil 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.

- 5.5.7.7 Equipment surfaces that regularly encounter 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).
- 5.5.7.8 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).
- 5.5.7.9 The bottom of benches for growing plants must be raised at least 46 cm above the floor surface to prevent contamination from splashing water.
- 5.5.7.10 Entry to the production site 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 production site. 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 area an area equipped with wash station(s), foot bath(s), and protective clothing (e.g., aprons, lab coats, gloves, see 5.6.4).
- 5.5.7.11 Place of production personnel must regularly scout the production site(s) for exposed soil, containment breaches, 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.
- 5.5.8 Packinghouse and Cold Room Infrastructure
 - 5.5.8.1 Packing area must be clean and disinfected before use.
 - 5.5.8.2 Packing and loading must take place inside a pest exclusionary structure.
 - 5.5.8.3 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.

- 5.5.8.4 All packing and shipping containers must be free of debris, soil, weeds, and pests.
- 5.5.8.5 Packing material must meet import requirements under Section 2-23 "Packing and Approved Packing Material" in the APHIS Plants for Planting Manual and the ACIR online database.
- 5.6 Sanitation Standards

5.6.1 All place of production personnel must wear protective clothing and follow the sanitation practices described in this REP to mitigate the risk of introducing regulated pests into production sites. The producer must strictly enforce sanitation practices in greenhouses, cold rooms, and grading or quality assessment facilities. Refer to APPENDIX 3 for a list of APHIS-approved disinfectants for surface, skin, and clothing.

5.6.2 The producer must ensure vehicles only enter the place of production or the clean production area(s) after their tires are cleaned and disinfected. The place of production may choose to prohibit them from entering the clean production area entirely.

5.6.3 No food is permitted in the greenhouses, cold rooms, and grading or quality assessment facilities.

5.6.4 Places of production must have a sanitation area for people entering each production site. Sanitation areas should include the following:

- 5.6.4.1 Wash Stations
 - 5.6.4.1.1 Places of production must have wash stations in production sites for use by each person entering the production site. All personnel must wash exposed body parts that may encounter plant material with soap or disinfectant prior to entering the production site.
 - 5.6.4.1.2 The use of gloves is optional. If used, personnel must disinfect or change gloves between each use or defined production unit, and between each production site. A use can be defined as work before or after a meal break, work from one bench to another, or between separations within the production site or other



identified spaces as identified by the place of production.

5.6.4.2 Footbaths

- 5.6.4.2.1 Places of production must have a sanitation area for footwear entering each production site. Places of production must provide footbaths to disinfect footwear as well as tools to brush or rinse footwear free of soil and debris. 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.
 - 5.6.4.2.2 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*. Place of production must change the footbath disinfectant a minimum of twice daily and remove any debris prior to replacement of the disinfectant.
 - 5.6.4.2.3 The place of production must maintain records at each footbath showing the disinfectant type and concentration used, and the date, time, name, and signature of personnel who serviced the footbath(s).

5.6.4.3 Protective Clothing area

- 5.6.4.3.1 All personnel entering production sites must wear protective clothing to prevent employee clothing worn outside the production sites contacting Rs R3bv2 host plants.
- 5.6.4.3.2 The place of production must provide production site-specific sets of protective clothing, except for shoes.
- 5.6.4.3.3 Personnel must only put on protective clothing after entering the sanitation area.



- 5.6.4.3.4 If personnel wear protective clothing between production sites, they must cover the used protective clothing with a clean layer of protective clothing.
- 5.6.4.3.5 The place of production 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.
- 5.6.4.3.6 The place of production must replace, disinfect, or wash protective clothing weekly. The unwashed or used protective clothing must be stored separately from clean protective clothing prior to washing or disinfection.

5.6.5 Personnel Hygiene

- 5.6.5.1 All personnel must follow the hygiene and sanitation standards in Section 5.6.
- 5.6.5.2 Personnel must regularly disinfect their hands and forearms or gloves by dipping or spraying with disinfectant after each defined production unit (e.g., every 10 plants, pots, or cuttings) and between plant varieties while working at the production site.

5.6.6 Tools and Equipment

- 5.6.6.1 Places of production must disinfect carts and other such equipment each day or when changing activities with an APHIS-approved disinfectant (refer to APPENDIX 3).
- 5.6.6.2 Places of production must disinfect all knives, scalpels, scissors, cutting collection containers, and other equipment that touch plants after each defined production unit (e.g., every 10 plants, pots, or 100 cuttings or bag) and between plant varieties and benches.
- 5.6.6.3 Places of production must disinfect hose ends that have been in contact with production site floors or other potentially contaminated surfaces prior to reuse.
- 5.6.6.4 The volume of disinfectant used to surface disinfect tools should be adequate to submerge the entire blade or other portions of tools that contact the plants. Disinfect for the minimum time



identified on the manufacturer label to be effective against *Ralstonia* solanacearum.

- 5.6.7 Handling of Cuttings and Traceability
 - 5.6.7.1 The place of production must demonstrate trace forward and traceback capability to a minimum of 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.
 - 5.6.7.2 The place of production must harvest cuttings for export directly into new plastic bags or into plastic containers that can be disinfected between each defined production unit.
 - 5.6.7.3 When transferring cuttings to grading or quality control production sites, bags or containers of cuttings must not touch any material that could expose them to U.S. regulated pests and disease.
- 5.6.8 Production Site Floors
 - 5.6.8.1 Production site floors and walkways must be cleaned at least weekly to remove debris and weeds. Production site personnel handling stock plants should not retrieve plant parts that fall to the floor (e.g., cuttings, trimmings) until after finished with pruning plants or harvesting activities. Plant debris collected in bags or bins should be removed from the production site daily.
 - 5.6.8.2 Producers must sanitize production site 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 REP requirements.
 - 5.6.8.3 Producers must retain production site cleaning records.
- 5.6.9 Growing Media
 - 5.6.9.1 All REP material must be grown in APHIS-approved growing media (refer to Table 7-1 in the APHIS Plants for Planting Manual).
 - 5.6.9.2 Growing media used for REP material must be stored in a manner that prevents contact with the ground, soil, or turf surfaces.
 - 5.6.9.3 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:



5.6.9.3.1 Steam pasteurization

- 5.6.9.3.1.1 Media must reach and uniformly hold a minimum temperature of 80° Celsius for two hours or 120 minutes.
- 5.6.9.3.1.2 A minimum of 10 sensors must be used to monitor temperature readings during treatment.
- 5.6.9.3.1.3 All sensors must reach a minimum of 80° Celsius before the twohour (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.
- 5.6.9.3.1.4 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.
- 5.6.9.3.1.5 Sensors must be evenly spaced on left, right, and center.
- 5.6.9.3.1.6 Sensors must be tested and calibrated before use. Calibrate temperature sensors in a swirling hot water bath with a factory calibrated, certified reference thermometer.
- 5.6.9.3.1.7 The certified reference thermometer must be a mercury, nonmercury, or digital thermometer with 0.1 $^{\circ}$ C (0.2 $^{\circ}$ F) graduations as a standard.
- 5.6.9.3.1.8 The temperature of the swirling hot water bath must consistently read the treatment temperature on the



certified reference thermometer. Place temperature sensors into the hot water bath and keep them there until the certified reference thermometer reads the treatment temperature for 10 consecutive minutes. After the temperature stabilizes remove the sensors and read the data.

- 5.6.9.3.1.9 Do not use any sensor that deviates by more than ± 0.3 °C (± 0.5 °F) from the treatment temperature. Record the greatest deviation for each sensor as the correction factor for that sensor. Any sensor that cannot be calibrated or repaired may not be used.
- 5.6.9.3.1.10 Places of production must keep calibration records.
- 5.6.9.3.1.11 Producers must retain growing media treatment records of the time, date, temperature, and duration of treatments. Records must show time, date, temperature, and duration of treatments. They must also include 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.

5.6.9.3.2 Fumigation

- 5.6.9.3.2.1 For methyl bromide the fumigation rate is 3 grams per liter of media for 72 hours at 21° Celsius or above.
- 5.6.9.3.2.2 For metam sodium 3% the applied concentration must be 50-ml per five-liter pot or bag that is kept covered for 72 hours.



5.6.9.3.2.3 Places of production must retain fumigation treatment records which include the time, date, temperature, fumigant concentration, and duration of treatment.

5.6.10 Plant Containers

- 5.6.10.1 Plant containers for all REP material must be disinfected prior to use and between callus batches or plantings.
- 5.6.10.2 Plant containers may be any of the following:

5.6.10.2.1	New plastic bags or pots.	

- 5.6.10.2.2 Recycled plastic pots that have been surface disinfected with an APHIS-approved disinfectant (APPENDIX 3).
- 5.6.10.2.3 Recycled plastic bags that have been sterilized along with reused growing media following requirements in Section 5.6.9.

5.6.11 Irrigation

- 5.6.11.1 All irrigation water must be free of Rs R3bv2. The water source, storage method, and irrigation system may require water treatment for Rs R3bv2.
 - 5.6.11.1.1 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.
 - 5.6.11.1.2 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 5.6.11.7).
- 5.6.11.2 Places of production must <u>not</u> irrigate plants using ebb and flow, overhead, or flood irrigation systems. Places of production may irrigate plants with overhead misting during callus production and initial establishment of new stock until they have established roots.

- 5.6.11.3 Irrigation systems must prevent emitters from contacting growing media or must include emitter backflow devices to prevent contamination of the watering system.
- 5.6.11.4 Places of production must hang watering hoses on a hook to prevent contact with the floor, production site 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 (refer to APPENDIX 3) if they touch the floor, plant material, or growing media.
- 5.6.11.5 Places of production 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.
- 5.6.11.6 To treat water, places of production must have a minimum of two independent water treatment systems: a primary system and a secondary back up system.
- 5.6.11.7 Each water treatment system must consist of <u>both</u> a filtration system and a purification system.
 - 5.6.11.7.1 Filtration systems must be:
 - 5.6.11.7.1.1 Reed-bed filtration,
 - 5.6.11.7.1.2 Slow sand filtration, or
 - 5.6.11.7.1.3 Ultra-filtration with a Molecular Weight Cut-Off (MWCO) of 150 kDa or less, and with proper filter maintenance as required by the manufacturer for proper functionality.
 - 5.6.11.7.2 Purification systems must be one of the following and must be monitored at regular intervals to ensure the system is functioning as required:
 - 5.6.11.7.2.1 Ozonation (0.4 ppm residual ozone for a minimum of four minutes).
 - 5.6.11.7.2.2 Ultraviolet irradiation:300J/m2 of UV light at 254 nm with at least 50% light transmission.



- 5.6.11.7.2.3 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.
- 5.6.11.7.2.4 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.
- 5.6.11.8 Places of production must keep water filtration and purification maintenance records. These include records verifying filtration and purification methods are operating within desired parameters (i.e., chlorine injections levels within required dosage, UV intensity maintained at required level; filtration efficacy).

6 Monitoring, Testing, and Sampling

6.1 Pest and Pathogen Monitoring

6.1.1 Each place of production must have a written pest management plan that covers REP standards. Additional pests or pathogens may be identified in the OWP depending on the country of origin. The plan must include the following minimum elements:

- 6.1.1.1 Identification of an employee as the designated place of production pest manager responsible for implementing the pest management plan.
- 6.1.1.2 Monthly structural inspection procedures to ensure compliance with minimum place of production standards (e.g., integrity of production site screening).
- 6.1.1.3 Procedures for the inspection of plant material before it enters the place of production.
- 6.1.1.4 Pest monitoring and control program procedures appropriate for the REP commodity and country of origin and to mitigate against the introduction of Rs R3bv2.



- 6.1.1.5 Procedures for plant pest and disease scouting, as well as removal and disposal of infested or infected plants. The plan should include the scouting schedule used to prevent or manage outbreaks, as well as the percentage of plants inspected per production site.
- 6.1.1.6 Traceability mechanisms used to facilitate traceback, and trace forward of REP commodities in the event of noncompliance or nonconformance.
- 6.1.1.7 Procedures for the inspection of plant material leaving the place of production.
- 6.1.1.8 Procedures to identify and report pathogens.
- 6.1.1.9 Procedures, documentation, and corrective actions following the detection of a regulated pest.
- 6.1.1.10 Procedures to control approved personnel and authorized visitor access to the place of production. Producers must maintain up-to-date lists of approved personnel.
- 6.1.1.11 Procedures for document control and retention. The producer must maintain records of pest monitoring and management activities.

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 The producer must maintain records of *Rs* testing and results.
 - 6.2.1.3 If any onsite testing is part of the place of production protocol, personnel conducting the tests must be trained in the method used and training records must be maintained by the place of production. If offsite testing is part of the place of production protocol, personnel must be trained in how to collect plant material to be sent for testing.
 - 6.2.1.4 APHIS-approved techniques for testing of routine samples (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. Places of production must maintain records of testing procedures,



Ralstonia Exclusion Program Framework

including the name of the test manufacturer, expiration date, and analytical sensitivity for tests other than PCR, qPCR or RPA.

- 6.2.1.5 APHIS-approved techniques for testing of symptomatic plant samples (see 6.2.3.2) are PCR, qPCR, or RPA.
- 6.2.1.6 Pooling samples for testing is permitted but must be preapproved. Producers must contact the NPPO and APHIS and submit the following information to the NPPO and APHIS for review and approval of pooling test procedures: testing technique and materials, how many samples per pooled test, and method for tracing pooled plants.
- 6.2.1.7 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 for testing must be taken from the lowest part of the stem that is above the soil even if the manufacturers' instructions state that leaves or other plant parts are acceptable.
- 6.2.1.8 When plant testing detects *Ralstonia solanacearum*, Rs R3bv2, or produces unexpected results, send the sample to an NPPO-approved laboratory for confirmatory testing.
- 6.2.1.9 Testing must be conducted weekly, with samples collected in accordance with ISPM 31 *Methodologies for sampling of consignments* (see Table 6-1).

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

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

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.



- 6.2.1.1 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.
- 6.2.1.2 Minimum sample size numbers are subject to change as new information regarding *Ralstonia* detection become available.
- 6.2.1.3 Facilities shall inform the NPPO of the exporting country immediately of any Ralstonia solanacearum or Rs R3bv2 detection while performing REP activities.
- 6.2.2 Elite Stock Testing
 - 6.2.2.1 All plant material used for REP 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 elite stock plant samples is PCR, qPCR, or RPA.
 - 6.2.2.3 Places of production must maintain documentation verifying imported elite stock, grafted stock or certified seed was tested and found free from Rs R3bv2 prior to shipment to the place of production.
- 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 <u>in addition</u> 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 production site 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 6.2.1.9).
- 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 production sites in the place of production (see ISPM 31, Section 3.1.3.3 *Stratified sampling* for further guidance). 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 production sites intended for export. The maximum number of samples that will need to be collected and tested per week for any lot size is 59 plants (ISPM 31 *Methodologies for Sampling of Consignments*; see Table 6-1, Section 6.2.1.9).
 - 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 nonsymptomatic plants. Non-symptomatic plants samples are not required to be destructively sampled.
- 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 APPENDIX 5).

6.3.2 Facilities shall inform the NPPO of the exporting country immediately of any *Ralstonia solanacearum* or Rs R3bv2 detection while performing REP 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 place of production and becoming established.

7.1.2 The Producer should limit production site access to personnel certified by the training.

7.1.3 The Producer must designate an employee as the training manager who is responsible for training all personnel in the following:

- 7.1.3.1 Proper production site 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 place of production.
- 7.1.3.4 Understanding how Rs R3bv2 can spread from plants outside the place of production 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, how personnel are certified, 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 place of production will result in the following immediate actions:

- 8.1.1.1 APHIS will issue an Emergency Action Notification (EAN).
- 8.1.1.2 APHIS will only allow the shipment to be reexported or destroyed.



- 8.1.1.3 APHIS will inform the NPPO and Producer of the detection, providing any associated information to enable traceback to the originating production site.
- 8.1.1.4 APHIS will initiate confirmatory testing and will not allow shipment of Rs R3bv2 host plants from the associated place of production to the United States while awaiting results.
- 8.1.1.5 APHIS will require trace forward information for the variety(ies) and production sites in the composite sample associated with the detection.
- 8.1.1.6 The place of production will be suspended following an Rs R3bv2 confirmation report from an NPPO-approved laboratory.
- 8.1.1.7 The NPPO and Producer must investigate the source of a confirmed detection, including a review of growing practices and increased scouting, sampling, and testing of plant material and the place of production's water source.
- 8.1.1.8 Before reinstatement of a suspended place of production, APHIS will communicate with the NPPO of the exporting country and the offshore place of production to discuss and agree on mitigation actions to reduce the risk of future *Rs* or Rs R3bv2 detections.

8.1.2 Any confirmed *Rs* or Rs R3bv2 detection found after consignments enter U.S. commerce that is traced back to a participating place of production will result in the following immediate actions:

- 8.1.2.1 The associated offshore place of production will be suspended following receipt of the Rs R3bv2 confirmed detection report from an NPPO-approved lab.
- 8.1.2.2 APHIS will notify the Offshore Producer by email of an *Rs* or Rs R3bv2 detection in the United States and provide traceback information and any results from confirmatory testing when available.
- 8.1.2.3 APHIS will require the offshore place of production to provide trace forward information for the variety(ies) and production sites associated with the detection.
- 8.1.2.4 APHIS will notify the NPPO through official communication.



- 8.1.2.5 The NPPO and Producer will investigate the source of the detection, including a review of growing practices within the place of production and increased scouting, sampling, and testing of plant material within and the place of production's water source.
- 8.1.2.6 Before reinstatement of a suspended place of production, APHIS will communicate with the NPPO of the exporting country and the offshore place of production to discuss and agree on mitigation actions to reduce the risk of future pest interceptions.

8.2 Non-conformance

8.2.1 Offshore places of production that do not conform with the REP's framework and associated bilateral agreement may not be certified/re-certified to participate in the REP. 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 place of production:

- 8.2.2.1 The place of production 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 The place of production 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 the offshore place of production to discuss and agree on mitigation actions to reduce the risk of future pest interceptions.
- 8.2.2.4 A confirmed detection following a recertification from suspension will result in immediate resuspension of the place of production from the REP for the rest of the shipping season, and until the next mandatory annual recertification audit.
- 8.2.2.5 Two confirmed detections within five years of a recertification from suspension results in suspension of the place of production in the REP until further notice.



- 8.2.2.6 Refer to APPENDIX 4 for detailed procedures following a confirmed Rs R3bv2 detection in an offshore place of production prior to export.
- 8.3 Oversight and Suspension

8.3.1 APHIS reserves the right to suspend the place of production from the REP (i.e., stop shipment of all exports to the United States) based on non-compliance or non-conformance issues.

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-CONFORMANCE AND OBSERVATIONS

Non-conformance Type: □ Critical □ Major	□ Minor
Description:	
Reference to Operational Work Plan/Framework	
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 issuance 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 place of production until <u>all</u> corrective actions are closed.

Immediate Corrective Action:
Corrective Action Documentation: □ Acceptable □ Not Acceptable □ N/A
Corrective Action is Acceptable: \Box Yes \Box 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:	□ Yes	□ No
APHIS Signature:		
Date Closed:		

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

1. FACILITY INFORMATION				
Facility Name			Date of	
Country			Inspection	
Country				· ~ \
Name & Address of Compan	y (as shown on Phyt	osanitary Certificate	e used for CBP Consignm	ent Clearance)
Is facility listed in the online	ACIR list at the	time of this		
audit?				
Finance Officer Email (at parel	t company headqua	rters)		
Physical Address of Inspecte	d Site			
GPS Coordinates of Inspecte	d Site			
Certified Facility Code				
Facility Manager Name:			Γ	
Email:			Phone:	
Trust Fund Manager (at	Name:			
company)			1	
Email:			Phone:	
Total Facility Area		Pelargonium	Area	
(all crops)	- 1	0		
Copy of facility map with to number of employees and	al			
layout of facility/greenhouse	5	Pelargonium	No. of Greenhous	ses
provided?				
Total No. of Greenhouses		No. Dolouro	uiuu at Easiliter	
(all crops)		No. Pelargor	nium at Facility	
	Total No. of Plants at Facility Greenhouses audited today (GH #)			I #)
(all crops)				
Total No. of employees		Greenhouses audited during last		
(peak season) inspection (GH #) U.S. Importer(s) Importer(s)				
Exporting Pelargonium cutti	ngs to the follow	wing countries:		

Dates of shipping season			
Dates of peak season			
What day(s) does the facility harvest/prepare cuttings for shipment?			
SUN MON TUE WED THU FRI SAT			
What day(s) of operation does the facility ship material to the US?			
SUN MON TUE WED THU FRI SAT			
Where does the facility source their nuclear stock/seed? (Provide facility name and physical address)			
Primary means of shipment Land Air Sea			
2. PLACE OF PRODUCTION INFRASTRUCTURE			
Entry to the main facility is secure and excludes any external source of <i>Ralstonia</i>			
solanacearum from entering the facility?			
Trucks entering the facility have tires cleaned and disinfected before entry into the			
production site OR are excluded from the production site?			
Disinfected Excluded			

USDA-APHIS Ralstonia Exclusion Program Framework

All Pelargon	ium are located wit	hin an enclosed greenhouse during all stages of growth?			
	There is complete separation between greenhouses?				
Employees w	Employees work exclusively in a block greenhouse during a single workday?				
	Plants are grown in a greenhouse unit that is used solely for <i>Pelargonium</i> grown in				
-	to USDA standard				
3. GREENHO	OUSE BUFFER ZON	E			
Sanitation	Buffer zone free of soil?	of grass, weeds, dicotyledonous plants, and exposed			
Size	There is at least a	There is at least a one-meter buffer zone around the entire greenhouse?			
Slope	Buffer zone is sloped away from the greenhouse or has canals to prevent water from entering the greenhouse?				
Slope		ruction is designed to prevent seasonal rain or flood ing the greenhouse?			
Composition		Rock Concrete Weed Cloth			
	Other:				
4. GREENHO	DUSE CONSTRUCTI				
	Тор	Glass Polycarbonate Polyethylene			
	- • P	Other:			
	Sides	Glass Polycarbonate Polyethylene Screen			
		Other: on or screen size (optional)			
Material	All vents and ope	nings in greenhouse are covered with screening to			
	-	of quarantine pests?			
5. ENTRY A	ND SANITATION ST				
	Single entrance in	to the greenhouse?			
	Direct access to v	estibule with wash station?			
Entry	Vestibule area wi	th closing double-door system?			
	Entry to production	on site restricted to authorized personnel?			
	Personal items stored before entry to greenhouse?				
	There is a footbat greenhouse?	h with rough bottom surface prior to entering the			
Footbath	Disinfectant/ concentration:				
	Volume covers so	les and lower portions of footwear?			
	Disinfectant is cha	anged 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/				
	Sink drains immediately to outside of production site?			
Disinfection Stations	Water source for			
Stations	sink: (Y or N next to selection)Sealed Well MunicipalOther:			
	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 worn by all personnel on entry to production site?			
	Types Washable lab coats Washable or disposable aprons			
	(circle)			
	washable or disposable aprons over lab coals			
	Other:			
	Protective clothing dedicated to each greenhouse and removed before exiting?			
Protective	If lab coats are worn between greenhouses, are they covered with or			
Clothing	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. GREENH	OUSE PRODUCTION SITE			
	Floors Gravel Crushed stone Concrete Other:			
	Walkways Gravel Crushed stone Concrete Other:			
Floors and Walkways	Is exposed soil present on floors or walkways?			
vv aikways	If yes, describe:			

	Debris-free? Weed-free?			
	Free of longstanding puddles of water?			
	If no, describe:			
	Floors and walkways are cleaned at least weekly. (i.e., swept, washed, brushed)			
	Describe:			
	during harvesti			
	Floors and wal	ways sanitized at least annually? How Often?		
	Disinfectant/ concentration:			
	Plants elevated	at least 46cm above greenhouse floor? How high?		
		the floor come in contact (e.g., splashing, watering) with		
	plants or bench			
Production		es (e.g., floors and benches) were sanitized prior to use?		
Surfaces	Disinfectant/			
	concentration:			
		ace design and composition ensures drainage?		
	Can irrigation water make pot to pot contact?			
	Ebb and flow irrigation system present?			
	Evidence of flood or sub-irrigation system? Emitters are separate from media in pots?			
		bed with backflow devices or raised above media?		
		y use overhead watering? If yes, describe:		
		y use overhead watering. If yes, describe.		
Irrigation System				
	Hose ends (last	1.2 meters) and irrigation nozzles are off greenhouse		
	floor or produc	ion surface?		
		de, hose and watering equipment treated with surface		
	disinfectant?			
	Disinfectant/			
	concentration			
Personal		oves and forearms disinfected by dipping or spraying with		
Hygiene	approved disin	ectant?		

	Disinfectant/	
	concentration:	
	Handa an alarra and disinfacted every they of Delana anion eviting a su	
	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?	
	Tools for harvesting (e.g., knives) soaked in disinfectant definable	
	production unit?	
	Production Unit	
	Tools for harvesting or processing <i>Pelargonium</i> cuttings (e.g., knives)	
Tools and	rotated between each stock plant or stock plant container/tray?	
Equipment	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	
	collection of harvested plants?	
Bags/	Plastic containers are disinfected before being reused?	
Containers	Disinfectant/	
	concentration:	
	Cuttings placed in plastic bags labeled with non-water-soluble ink?	
	Labels accompany each bag/container of cuttings?	
	Label system allows trace forward through rooting stations or directly	
	from farm to first wholesale customer?	
Cuttings	Label system allow traceback to the production site where they	
	originated?	
	Cutting containers do not contact soil or other material harboring	
	<i>Ralstonia</i> when transferred to grading facilities or cold room?	
Grading/	<u>Grading / Quality control</u> (circle one) is conducted during harvest at the	
Quality	bench?	
Zumity		

Control/	a.) Is Grading / Quality control (circle one) conducted in the					
Packing	greenhouse at an inspection table?					
facility	b.) Is Grading / Quality control (circle one) conducted in a separate					
	room?					
	Grading/Ouality control	l facilities satisfy same conditio	ns as greenhouses with			
	respect to:					
	Wash Stations Hand Washing Footbath					
	Protective		T 1			
	Clothing	Personal Hygiene	Tools			
	Handling of	Floors	Production			
	production stock	FIOOIS	Surfaces			
	Water Treatment	Training Personnel				
		surface disinfected between bag	s or containers of			
	cuttings processed.					
7. GROWIN						
		PHIS-approved (e.g., Scoria, Vo	lcanic rock,			
		ix, describe composition)?				
	Describe:					
Туре						
	If no, describe:					
Safeguarding	Is media and/or pots stored on soil/dirt or turf surfaces?					
	Is media sterilized?					
	Method of treatment (Enter Yes or No for relevant treatments)					
	Steam: tw	o hours after all ten sensors read	ch 80°C			
			701 (> 70°E)			
	Fumigatio	on: Methyl Bromide (3g/liter for	$72n (\underline{w} \ge 70 \text{ F})$			
Sterilization	Fumigatic	n: Metam Sodium (e.g., Vapam	3% (equivalent to 50 ml per			
	Fumigation: Metam Sodium (e.g., Vapam, 3% (equivalent to 50 ml p bag covered for 72 hours)					
	6					
	Is media fumigated on a non-porous surface?					
	Time, temperature, and duration of treatment at all sensors is recorded/logged for growing media treatment? (records maintained for three years)					
Containers	Select One: Plastic pots Plastic bags Are pots/bags new each year?					

	If no, describe below:			
8. WATER T	REATMENT			
	Municipal	Deep, Sealed Well		
	Other:	- <u>-</u>		
	If "other," describe and in	dicate treatment:		
			-	
		ge such that there is no opportunity	for water to	
	be contaminated by soil of			
		Itration and/or slow sand filtration s		
		combination with one of the following		
	Ozonation (0.4 ppm residual ozone for \geq 4 minutes.)			
	Ultraviolet irradiation (300J/m ² UV @ 254 nm, \ge 50%			
	$\frac{1}{1}$			
	Peroxygen products (residual level ≥ 4 mg / liter per acetic acid for 2 min.) (Achieved at injection 15-35m ³ /hour with concentration of			
		ml/m3 of peracetic acid.)		
Source		the dioxide (residual level $\geq 0.1 \text{ mg}$)	/ liter chlorine dioxide	
		min.) (Achieved with 5mg/L inject		
		O_2 generator.)	0	
		iltration (per particle size.)		
	Independent backup water treatment system in place in case primary system fails?			
	Is there a log of any routir	ne inspection, breaches, or maintena	ance issues in	
	the irrigation system kept			
		east twice a year to ensure freedom	from	
	Ralstonia solanacearum?			
	Log of testing kept on site			
	Detailed description of water treatment (use additional pages if necessary):			
9. PEST SUF	RVEY AND DETECTION			
Testing	What testing methods, if a	nny, are used (e.g., ELISA, PCR, ap	proved lateral flow and	
Methods	strip serological test kits,		-	
		1 0/		

	Describe:				
	Testa esa dueta det e				
	Tests conducted at a	pproved/certified:	NIDDO		
	In-house laboratory: NPPO laboratory:				
	Third party laborator	ry:	Other:	I	
	Tests conducted by, or under the supervision of, the plant protection				
	-	ountry of origin or their de			
	Records available fo	r inspection (a minimum o	of three years)?		
	Are samples collecte	ed at a rate consistent with	IPPC ISPM 31?		
	Describe sampling p	rogram:			
	Sampling done prior to first shipment and weekly throughout shipping				
	season?				
	Approved lateral flow and strip serological test conducted correctly?				
	Which plant tissue is tested for <i>R. solanacearum</i> ?				
	All suspect wilted plants must be sampled with root crown tissue. Pathogen concentrates				
Sampling	in the lower stem. Testing leaves and partial stem samples is not acceptable.				
	Plants scouted regularly for signs of wilt and tested as necessary?				
	Test performed prior to destruction of discarded plants (except when 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? (a minimum of three years)				
	Production stages sa	mpled:			
	Nuclear/Foundatio		Sampling R	late:	
	n stock		1 0		
	Increase block		Sampling R		
	Production block		Sampling R	late:	
10. TRAININ	1	1 1 1 -			
Employee	Number of Employees during peak season?				
Training	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) Annual Monthly Quarterly Weekly Bi-weekly
	Annual Monthly Quarterly Weekly Bi-weekly Describe:
	Describe.
	A list of trained personnel is maintained? (a minimum of three years)
	List the last date of training conducted:
	Access to greenhouses is limited to trained and certified individuals?
	Facility maintains record of training of personnel and certifications?
1. SUIVIIVIAI	RY, CORRECTIVE ACTION REQUESTS and FOLLOW-UP
Observations and	
and	
Summary	
(Attach additional	
pages, if	
necessary)	

	Please list deficiencies noted, recommendations for resolution, corrective actions, and agreement on follow up (complete Corrective Action Request (CAR)), if available:
Corrective Actions	
and	
Follow-up	

I have reviewed and agreed with the above and will immediately resolve outstanding issues detected:

APHIS Inspector:	Signature:	Email:	Date:
NPPO Inspector:	Signature:	Email:	Date:
Facility Representative:	Signature:	Email:	Date:

APPENDIX 3: LIST OF APPROVED DISINFECTANTS

Table A3-1: Examples of APHIS-approved Surface Disinfectants.¹ Any product used for disinfection should be approved by the NPPO, labeled for use by the proper authority in the country where the place of production 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.
 - A3.1.1. Any product used for disinfection should be approved by the NPPO, labeled for use by the proper authority in the country where the place of production is located, and must be efficacious against plant pathogens. (Last updated 2007).
 - A3.1.1.1. Alcohols (>60% ethanol)
 - A3.1.1.2. Chlorhexidine (0.5%-4% depending on preparation)
 - A3.1.1.3. Chloroxylenol (0.3%-3.75%)
 - 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 place of production 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 PLACE OF PRODUCTION

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 and unexpected results, the place of production must send a sample to an NPPO-approved laboratory for Rs R3bv2 confirmatory testing using an APHIS-approved 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 REP 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:
 - A4.1.4.1. Stop shipping all REP material to the United States until further notice.
 - A4.1.4.2. Contact APHIS by email or phone within 24 hours and provide variety and production site 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.
 - A4.1.4.3.1. 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.
 - A4.1.4.4. Supply APHIS consignment data for all shipments from the place of production 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; and
- A4.1.4.4.6. 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 were 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 REP requirements or other identified gaps, 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 place of production, 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 place of production, production site(s), and varieties:
 - A4.2.2.1. immediately if there are shipments in transit from the associated place of production, 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 place of production 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 recertify the suspended place of production before adding 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.

USDA-APHIS Ralstonia Exclusion Program Framework

- A4.3.9. Will enforce consequences of additional Rs R3bv2 detections:
 - A4.3.9.1. Additional Rs detections following recertification of a suspended 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 following a recertification from suspension will result in immediate resuspension of the place of production from the REP for the rest of the shipping season, and until the next mandatory annual recertification audit.
 - A4.3.9.3. Two confirmed Rs R3bv2 detections within five years of recertification from suspension 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 PLACES OF PRODUCTION**

Participating places of production must test their water source, treated irrigation water, and recycled water after treatment for *Ralstonia solanacearum* at least every six months. Testing irrigation runoff water for *Ralstonia solanacearum* is not required, however we supply recommendations in A5.2.

Water samples must be sent to an NPPO-approved laboratory. Places of production must use one of the APHIS-approved methods below to test for *Ralstonia solanacearum* in water:

- A culture-based method combined with molecular identification, or
- A filtration-based method coupled with molecular detection.
- A5.1. Detection Methods for Ralstonia solanacearum in Treated Irrigation Water
 - A5.1.1. Water must be collected from holding tanks using sterile tubes or bottles.
 - A5.1.2. Collect the volume of water required by the chosen testing method.
 - A5.1.3. Duplicate samples must be tested. Samples should be transported in cool, dark conditions (4–10°C) and tested within 24 hours.
 - A5.1.4. If using a culture-based approach combined with molecular identification tests, places of production should follow the EPPO Bulletin 48 standard for testing water.
 - A5.1.4.1. Concentrate collected water samples using one of the following methods:
 - A5.1.4.1.1. Centrifugation of 30–50 mL sub-samples followed by resuspension of the pellet.
 - A5.1.4.1.2. Membrane filtration (1 L through a maximum pore size of 0.45 μm) followed by washing of the membrane.
 - A5.1.4.2. Perform dilution plating of the concentrated samples on a semi-selective SMSA/Sequeira medium. Test for typical colonies using PCR as specified in EPPO Bulletin 48.
 - A5.1.5. If using a filtration-based method coupled with molecular detection
 - A5.1.5.1. Places of production should follow the APHIS Plant Pathogen Confirmatory Diagnostic Lab (PPCDL) protocol for testing water. Please refer to the APHIS REP website for instructions on obtaining the PPCDL protocol. The protocol may be updated at any time. Places of production are responsible for using the most updated version of the water testing protocol found on the APHIS REP website.

- A5.1.5.2. 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 1×10^1 CFU/ml.
- A5.2. Recommendations for testing run-off water
 - A5.2.1. Places of production may choose to recycle irrigated run-off water in an effort to conserve resources and reduce 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 production site level. Places of production must report Rs R3bv2 detections to the NPPO.
 - A5.2.2.2. While recommended for monitoring purposes, runoff water testing is not included in the REP requirements.
 - 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) and the effectiveness of the water treatment process.
 - A5.2.4. Inhibitory substances must be neutralized prior to testing. The 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.

Note: Treated run-off water that will be used for irrigation must be tested as described in A5.1.

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