Survey and Response Plan

Tobamovirus: Tomato Brown Rugose Fruit Virus (ToBRFV)

Cover Image. Infected tomato fruits can be rough, deformed, brown, necrotic, and wrinkled, while leaves are mottled or mosaic (image credit José Antonio Garzón-Tiznado, Universidad Autónoma de Sinaloa)

This survey and response plan provides measures necessary to contain and eradicate ToBRFV from U.S. greenhouse facilities. This plan supplements information contained in the current ToBRFV New Pest Response Guidelines (NPRG). Official should use both the plan and NPRG when conducting surveys and regulatory actions associated with ToBRFV.
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**Introduction**

Tomato brown rugose fruit virus (ToBRFV) is a virus of tomatoes, peppers, eggplant, and other solanaceous hosts ([Refer to the Pest Overview section of the NPRG for a full list of hosts](#)). The virus is transmitted in seed and spreads easily when healthy plants come in contact with contaminated equipment, hands, clothing, or infected plants or plant parts. Because this virus is difficult to control and eradicate, prevention and early detection are critical to safeguard U.S. agriculture.

Plant Protection and Quarantine (PPQ) officials and cooperators conducting PPQ-funded solanaceous surveys should include ToBRFV in commercial greenhouses. The focus on greenhouse production rather than field production is in part due to resource limitation as well as the horticultural characteristics inherent to greenhouse systems. Given the dense plant spacing, frequent handling and manipulation of host material, there is increased likelihood of mechanical spread and the potential for higher incidence of infection in greenhouses.

Officials must collect symptomatic samples to conduct immunostrip testing or submit to a screening laboratory for diagnostic testing. Presumptive positive samples must be forwarded to PPQ’s Beltsville Laboratory for confirmatory testing.

Upon PPQ’s confirmation of a ToBRFV-positive sample, Federal and State officials must take regulatory action to remove and properly dispose of all infected plant material and reduce the risk of spreading ToBRFV within the affected greenhouse or nursery. Officials should conduct a delimitation survey to delineate the core and buffer areas within the greenhouse. In addition, officials should work with the greenhouse to identify the least restrictive action for the remainder of the facility, depending on the cultural and sanitation practices that are in place.

**Survey Preparation, Sanitization and Clean-Up**

When conducting detection or delimitation surveys, officials must follow strict protocols for preventing contamination between greenhouses, nurseries, and other operational facilities. Two basic principles should govern surveys and the sampling process:

1. **ToBRFV cannot be diagnosed through the visual inspection of symptoms alone; only laboratory testing can provide a definitive diagnosis.**
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2. ToBRFV is easily spread by mechanical transmission; take all recommended precautions to ensure you do not inadvertently contribute to its spread.

Officials should bring disposable gloves, disposable protective clothing, small zip-lock bags, permanent markers, heavy Styrofoam containers of different sizes, a few old newspapers, shipping tape, several frozen ice packs, and a laptop with internet capability. Officials should download PPQ Form 391 to their mobile data collection device or bring physical copies of the form to the location.

For all site visits, officials must take the following precautions:

1. Before starting a survey, determine whether the facility has recently applied any pesticides that would render it unsafe to inspect the plants. Contact the property owner or manager and ask if there is a re-entry period in effect due to pesticide application. Look for posted signs indicating recent pesticide applications.

2. Determine whether quarantines for other pests or crops are in effect in the survey area. Comply with all quarantine requirements.

3. Take strict measures to prevent contamination between properties during inspections.
   a. Designate a clean area where transport vehicles can park.
   b. Use disposable protective clothing, gloves, and footwear, and change them before entering each greenhouse unit.
   c. Clean and sanitize equipment, tools, and footwear after each use.
   d. Disinfect used tool(s) with any of the following EPA approved disinfectants:
      ● 10% bleach solution: 1 part bleach (any commercial bleach) to 9 parts water
      ● 20.4% potassium peroxymonosulfate, 1.5% sodium chloride applied at 2.0% (20 g/L or 0.17 lb/gal)
      ● 20% solution (wt/vol) of non-fat dry milk plus 0.1% polysorbate nonionic surfactant
   e. Thoroughly spray tools with or immerse in the disinfectant and let them air-dry.
   f. Change gloves after touching an infected or suspected infected plant.
   g. Disinfect vehicles and large equipment (e.g., storage areas and bins).
Detection Survey

Purpose

This section outlines the protocol for conducting ToBRFV detection surveys on seedling and fruit hosts in commercial greenhouses. Detection surveys are designed to determine whether ToBRFV is present or absent in a defined area.

Procedure

1. Information Collection System

Enter data into the approved data collection system. Officials should also follow their cooperative agreements for data requirements.

2. Conduct a Visual Survey

Visually examine host plants and fruit for symptoms of ToBRFV. Refer to the Pest Identification section of the NPRG for signs and symptoms. Since ToBRFV is mechanically transmitted, visually inspect areas where equipment and people are most likely to be in contact with plant material, especially in entry and exit areas. Inspect greenhouse waste areas for discarded plants. Ask the greenhouse owner or manager to identify cull piles. Examine them for the presence of recently discarded or dead host plants.

Officials should use the following decision table before starting the leaf sample collection.

<table>
<thead>
<tr>
<th>If host plant is:</th>
<th>And:</th>
<th>And:</th>
<th>Then:</th>
</tr>
</thead>
<tbody>
<tr>
<td>Showing symptoms</td>
<td>Plants are at the flowering or fruiting stage</td>
<td>Surveyor is performing immunostrip testing</td>
<td>Issue an EAN and follow the procedures in ‘Conducting Agdia Immunostrip Testing’ and ‘Preparing Samples for Shipment to Beltsville’</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Surveyor is not performing immunostrip testing</td>
<td>Issue an EAN and follow the procedures for ‘Preparing Samples for Shipment to Screening Labs’</td>
</tr>
<tr>
<td></td>
<td>Plants are at the seedling stage with multiple sets of true leaves but not yet flowering or fruiting</td>
<td></td>
<td>Issue and EAN and follow the procedures for ‘Preparing Samples for Shipment to Screening Labs’</td>
</tr>
</tbody>
</table>
### 3. Issue an Emergency Action Notification (PPQ Form 523, EAN) and/or Comparable State Notification to the Owner or Operator of Each Property Where TOBRFV is Suspected

If host plants exhibit symptoms of ToBRFV, issue an Emergency Action Notification (EAN) and place the facility on hold, pending confirmation of samples. Recipients of EANs are required to implement treatments and handling procedures. A PPQ Plant Health Safeguarding Specialist (PHSS) or a State inspector must witness, supervise, and document those activities.

Issue EANs and/or comparable State notifications to the property owners or managers of all establishments that are involved in the handling, moving, or processing of articles capable of spreading ToBRFV. You should issue an EAN pending positive identification. EANs are issued under the authority given in 7 CFR 330. **States should issue their own hold orders parallel to the EAN to prevent the intrastate movement of plants.**

The following language should be included on the EAN:

> “All [insert host: tomatoes, eggplants, peppers] (plants, seed, seedlings) are prohibited from movement from the nursery property pending further notification by USDA–APHIS–PPQ. All other host plants are also subject to this hold and must be reported immediately to USDA–APHIS–PPQ and held until further notice. The above listed plants and all potentially infested material, potting soil, and plant containers, may be destroyed in accordance with USDA policies. Areas housing infected material, irrigation systems, irrigation ponds, outdoor soil or holding areas in association with infected material, and contaminated equipment will be cleaned and decontaminated according to USDA protocols.”

### 4. Greenhouse Leaf Sample Collection

If host plants exhibit symptoms of ToBRFV, follow the sampling protocol below to collect, test, and/or submit samples.

Each greenhouse is considered a sampling unit. Sample the greenhouse by collecting symptomatic host plant leaves.

**Conducting Agdia Immunostrip Testing (If official is not conducting the testing on site or symptomatic plants are at the seedling stage, see “Preparing samples for Shipment to National Plant Diagnostic Network” below.)**

A. Wear gloves when collecting samples.
B. Collect five to seven young leaves from each symptomatic plant, up to a maximum of five samples for each host variety, or a maximum of 10 samples total. (Note: this applies only to symptomatic plants that are at the flowering or fruiting stage.)
C. )
1. Invert a plastic re-sealable bag over your hand and use it to remove five to seven symptomatic young leaves from the plant.
2. Visibly and indelibly flag or mark plants and areas sampled.
3. Change gloves and disinfect any tools that may have touched the plant.
4. Label the outside of each bag with the following:
   i. A unique identification number and the date
   ii. Collector’s name
   iii. Specific location in the greenhouse
D. Remove one leaf (select the most symptomatic leaf of the approximate size) of approximately 1 square inch (the size of a quarter) and place inside the mesh bag provided in the Agdia kit.

E. Perform the immunostrip test following the manufacturer’s instructions. If the control line does not appear on the immunostrip:
   1. Make sure there are no bubbles or frothing inside the bag; flick the bag to break bubbles.
   2. Re-use the same bag containing buffer and plant material and test again with a new immunostrip.
   3. Confirm that the control line is present with the new strip.
   4. If the control line fails to appear a second time, select another leaf and repeat the steps using a fresh kit.
F. If the immunostrip is:
   - Negative – no further action; discard remainder of plant material using the facility’s regular methods for plant disposal. Remove the facility from the EAN if all samples are negative.
   - Positive – follow additional steps below. All the remaining tissue from any presumptive positive samples must be sent to PPQ’s Beltsville laboratory for confirmatory testing.

Preparing Samples for Shipment to Beltsville
A. Fill out a PPQ Form 391 for each sample. Place up to 10 individual bags into a larger, 1-gallon plastic re-sealable bag.
B. Place a hard copy of each completed PPQ Form 391 inside the outer bag. Each PPQ Form 391 should include the corresponding identification numbers for the individual bags.
C. Keep plant tissue refrigerated until shipment; do not freeze samples. **Send samples as soon as possible.**

D. Place samples into a heavy Styrofoam container with a few ice packs. Do not place the samples directly on the ice packs. Use newspaper or other materials to prevent direct contact.

E. Seal all seams of the shipping container with shipping tape.

F. Prior to shipping samples, submitters should contact PPQ’s Beltsville Laboratory and the Domestic Diagnostic Coordinator by email:

   APHIS-PPQCPHSTBeltsvilleSampleDiagnostics@usda.gov;
PQQ.Diagnostic.Coordinator@usda.gov

In the email, include the tracking number and the number of samples being and attach an electronic copy of PPQ Form 391(s).

G. Ship via overnight express courier (FedEx, UPS, etc.) Monday through Thursday to PPQ’s Beltsville Laboratory at the following address:

   Sample Diagnostics  
   USDA–APHIS–PPQ–S&T  
   Building580, BARC-East  
   Powder Mill Road  
   Beltsville, MD 20705-2350  
   Phone: (301) 313-9208

**Preparing Samples for Shipment to National Plant Diagnostic Network (NPDN) Screening Labs (If official is not conducting immunostrip testing on site)**

A. Wear gloves when collecting samples.

B. Collect five to seven young leaves from each symptomatic plant, up to a maximum of five samples for each host variety, or a maximum of 10 samples total. (Note: If the plants are very young and do not yet have multiple sets of true leaves, do not sample and return to the facility when the plants have matured and have enough material to collect.)

1. Invert a plastic re-sealable bag over your hand and use it to remove five to seven symptomatic young leaves from the plant. Do not include any soil media inside the bags.

2. Visibly and indelibly flag or mark plants and areas sampled. Do not remove leaves from the plastic re-sealable bag after it has been sealed.

3. Change gloves and disinfect any tools that may have touched the plant.

4. Label the outside of each bag with the following:
   i. A unique identification number and the date
   ii. Collector’s name
   iii. Specific location in the greenhouse

5. Place up to 10 individual bags into a larger, 1-gallon plastic re-sealable bag.
   i. Place a hard copy of the completed PPQ Form 391 inside the outer bag. Each PPQ Form 391 should include the corresponding identification numbers for the
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individual bags.

C. Keep plant tissue refrigerated until shipment; do not freeze. **Send samples as soon as possible.**

D. Submit samples to an NPDN screening laboratory.

E. Prior to shipping samples, submitters should contact the screening lab via phone or email to ensure they are available to receive the samples.

F. Seal all seams of the shipping container with shipping tape.

G. Ship via overnight express courier (FedEx, UPS, etc.) Monday through Thursday. Do not ship on Fridays.

H. Any sample(s) testing positive in a screening lab must be submitted to PPQ’s Beltsville laboratory for confirmation.
Section 3

Regulatory Procedures Following the Confirmation of ToBRFV

Purpose
This section outlines the regulatory procedures following the confirmation of ToBRFV in a greenhouse.

Procedure

1. Notify the Grower
In the event of a positive confirmation from the Beltsville Laboratory, the State Plant Regulatory Official (SPRO) or State Plant Health Director (SPHD) should immediately contact the facility owner to inform them of the confirmed positive sample and ensure a hold has been placed on all regulated plants (intended for fruit and as seedlings). Arrive on site as soon as possible.

2. Interview Grower and Review and Document Information

Ask about Cultural and Sanitation Practices

A. Does the irrigation system use recycled water? If not, does it have back flow prevention?
B. Does the facility use hydroponics?
C. Where are cull piles and compost located?
   - Cull and compost piles may be found next to greenhouses, on other farm property areas, or near storage facilities or processors. These are potential reservoirs of the pathogen and are regulated until the risk level is determined by investigations and/or testing.
D. What sanitation practices are used for agricultural machinery, equipment, and tools?
   - Harvesting equipment including trucks, harvesters, trailers, machinery, tools, boxes, worker footwear or implements, transport conveyances, and storage facilities are subject to cleaning and disinfection if used in a greenhouse where ToBRFV was detected. Tools, implements, equipment, benches, and greenhouses used in cultivation that may have contacted positive tested plant material are subject to disinfection.
E. What sanitation practices do employees use when working with plant material (seeds, seedlings, plants, soil, cull piles, compost, irrigation water, backflow prevention)?
F. What is the composition of benches and flooring?
Ask for Trace Information

A. What is the origin of the seed, grafts, or transplants?
B. What is the history of movement of the plant material within the facility (example: where are they rooted or grafted)?
C. Have any products moved out of the facility and where have they gone?

Review Documents

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

For positive detections in seed lots, rely on shipping records and information provided by the grower, farm manager or facility manager for how many plants or seed remain, how plant material and seed have moved within the facility, destination of material sold, origin of seed, and cultural and sanitation practices employed.

Record Information

Keep a detailed accounting of the numbers and types of plant material held and/or destroyed in control actions. Consult a master list of varieties distributed with the lists of facilities. Draw maps of the facility layout to locate suspect plants, other potentially infected lots, cull piles, and water runoff areas. Take photographs of the facility layout, watering and sanitation methods, materials and methods used, labeling, and any other situation that may be useful for documentation and analysis. Store all records in the centralized location.

3. Conduct Trace Work

Use information from grower documentation and interview to:

A. Trace forward any suspected positive seed lots, rootstock, scions, or plants to see where else they may have been planted.
B. Trace back any suspected positive seed lots, rootstock, scions, or plants to identify the infection source of confirmed positive ToBRFV plants.
C. If trace work involves seed, refer to the Delimitation Survey section of the NPRG for guidance on sampling seeds.

Trace work actions

<table>
<thead>
<tr>
<th>IF</th>
<th>THEN</th>
</tr>
</thead>
<tbody>
<tr>
<td>Traced to seed lot</td>
<td>Continue trace work to source of seeds. Survey seed lot and collect records regarding other locations that received the seed lot. Trace work must continue to locations that received the seed lot and surveys must be conducted at those locations.</td>
</tr>
<tr>
<td>Traced to fruit in market</td>
<td>End response due to likelihood fruit will be consumed prior to regulatory action.</td>
</tr>
</tbody>
</table>
4. **Identify and Apply the Appropriate Mitigation Option**

At this time, no chemical controls are available to treat ToBRFV-infected plant material.

**Seed**

Seed producers must destroy and/or dispose of all positive seed lots in their entirety (refer to **Option 2: Host Removal**). We recommend testing all remaining lots in the inventory for the presence of ToBRFV. Refer to **Delimitation Survey section of the NPRG for guidance on sampling seeds**. Seed producers must further clean and disinfect all equipment, storage areas, and conveyances used in their business enterprise that may have come in contact with the positive lot(s). They must also make regular testing for ToBRFV a part of their seed quality management program.

**Seedlings and Plants**

**Option 1: Partial Host Removal**

Prior to following the protocol for Option 2, a delimiting survey is required. Refer to the **Delimitation Survey section of the NPRG for guidance**.

In consultation with regulatory officials, an owner may elect to do partial removal in an attempt to eliminate the disease.

If this option is chosen, the following steps must be completed in sequence and verified by a regulatory official:

- **A.** All plants confirmed positive following the detection survey must be destroyed along with plants in the two rows on either side of the confirmed positive(s). A total of 5 rows must be destroyed following the destruction procedure in **Option 2** below.
- **B.** Begin delimiting survey to establish the core infested area. Refer to the **Delimitation Survey section of the NPRG for guidance**.
- **C.** Once the core infested area is defined (no more positive plants), establish the buffer area as per Survey Techniques for Delimitation.
- **D.** Once the buffer area is established (3 times the diameter of the core infested area), destroy all plant material within the buffer area.
- **E.** Quarantine the remaining rows of the greenhouse, monitor plants at least weekly for symptoms, and randomly sample plants for infection for at least 30 days.

**Option 2: Full Host Removal**

All host plant material must be removed and properly disposed of using PPQ-approved disposal methods. Destroy or dispose of all plant material that can be reasonably removed. This includes cull piles and other plant debris. Apply a heat treatment (steam/autoclave) with an internal...
temperature of 100 °C (212 °F) for a minimum of 30 minutes to all plant material (i.e. plant parts, soil) that may contain or is suspected of containing ToBRFV. Bury all material in a landfill. Because the heat produced within compost piles is not sufficient to deactivate ToBRFV, use the same heat treatment as stated above or incinerate the compost pile if there is evidence of infected material.

Alternatively, plant material, associated growth media, associated containers including pots and trays, all leaf debris in and around the area where plants were stored can be double bagged using plastic bags of 2 mil thickness or greater and buried to a depth of no less than 6 feet (1.83m). The material must be buried at onsite, at a USDA-approved site, or municipal landfill, which is expected to remain undisturbed. Make every effort to prevent plant debris or soil from being dislodged from the plants. The burial site must be safeguarded to prevent removal of quarantine material. All of these actions must be verified by a PPQ PHSS or a State inspector.

5. **Ensure all Appropriate Equipment and Surfaces are Disinfected**

**Tractors and Large Machinery**

Pressure wash (with detergent) equipment, such as tractors or other large machinery, that may have come in contact with infected plant material. Follow the two-step process below:

1. Power wash or steam clean equipment to remove all soil and debris before it leaves infected areas; AND
2. Disinfect all equipment (see below).

**Other Equipment**

Use one of the following EPA approved disinfectants, with an exposure period of at least 15 minutes, on anything (e.g. tools, plant trays, benches, drains, water storage areas, irrigation systems) that may have come into contact with infected plants and plant material:

- 10% bleach solution: 1 part bleach (any commercial bleach) to 9 parts water
- Use 20.4% potassium peroxymonosulfate, 1.5% sodium chloride applied at 2.0% (20 g/L or 0.17 lb/gal)
- 20% solution (wt/vol) of non-fat dry milk plus 0.1% polysorbate nonionic surfactant

6. **Witness all Actions**

All actions related to destruction and disinfection must be witnessed, supervised, and documented by a PPQ PHSS or a State cooperating inspector.

7. **Provide Guidance**

Provide the grower with the *Strategies for Preventing the Introduction and Spread of ToBRFV* document (see Appendix A) that outlines how to prevent the spread of ToBRFV to uninfected areas and avoid future introductions of the virus.
8. Release from EAN

A property may be released from an EAN when the following conditions can be demonstrated:

- No suspect plants or seed from specified distribution facilities were received at the destination during the specified time periods.
- No other host plants from the farm have tested positive or shown ToBRFV symptoms. If symptoms were detected, the plants were subsequently sampled, tested, and found negative for ToBRFV.
- Suspect seed or greenhouses with confirmed detections of ToBRFV were mitigated with approved methods. This includes removal and destruction of hosts and the disinfection areas where plants were destroyed, including the tools and other equipment that may have come in contact with potentially contaminated plants. This is sometimes referred to as destruction in lieu of identification.
Appendix A

Strategies for Preventing the Introduction and Spread of Tomato Brown Rugose Fruit Virus

The following document provides guidance to growers on how to prevent spread of ToBRFV and to prevent future introductions of ToBRFV.
Strategies for Preventing the Introduction and Spread of Tomato Brown Rugose Fruit Virus

Overview

Tomato brown rugose fruit virus (ToBRFV) is a virus of tomatoes and peppers. The virus is easily spread when healthy plants come in contact with contaminated equipment, hands, clothing, or infected plants or plant parts. Because the virus is difficult to control or eradicate, prevention is critical. This document details strategies to reduce your operation’s likelihood of exposure to ToBRFV. It also offers strategies for responding to ToBRFV detections, reducing spread in facilities, and preventing reinfection.

Preventing the Introduction of ToBRFV

To reduce the risk of introducing ToBRFV into your operation, use the following sanitation and preventive practices:

- Use seed, seedling, and graft material that are certified virus-free.
- Minimize crop handling and other procedures that may wound the plants, especially early in the growing season. Early infections may result in the most severe yield losses.
- Treat each greenhouse as a separate unit, and follow the sanitation measures listed on page 3.
- Report any signs or symptoms of this disease to your State Plant Regulatory Official or to your local USDA Plant Protection and Quarantine Office.
Response Strategies if ToBRFV is Detected

Treatment Measures

At this time there are no chemical controls available to treat ToBRFV infected plants. Remove all infected plant material from the greenhouse—including cull piles and other plant debris—and use the following APHIS-approved disposal method:

- Apply a heat treatment (steam/autoclave) with an internal temperature of 100 °C (212 °F) for a minimum of 30 minutes to all plant material (i.e. plant parts, soil) that may contain ToBRFV. After completing the heat treatment, the material can be buried in a landfill. Use the same treatment for compost piles or incinerate.

OR

- Double bag all Infected plants, associated growth media, associated containers including pots and trays, all leaf debris in and around the area where plants were stored using plastic bags of 2 mil thickness or greater. Bury to a depth of no less than 6 feet (1.83m). The material must be buried onsite, at a USDA-approved site, or municipal landfill, which is expected to remain undisturbed. Make every effort to prevent plant debris or soil from being dislodged from the plants.

Disinfection Measures

- Pressure wash (with detergent) large machinery or equipment (scissor lifts, etc.), that may have come in contact with infected plant material. Follow the two-step process below:
  1. Power wash or steam clean equipment to remove all soil and debris before it leaves infected areas; AND
  2. Disinfect all equipment (see below).

- Use one of the following disinfectants, with an exposure period of at least 15 minutes, on anything that may have come into contact with infected plants and plant material:
  - 10% bleach solution: 1 part bleach (any commercial bleach) to 9 parts water
  - Use 20.4% potassium peroxymonosulfate, 1.5% sodium chloride applied at 2.0% (20 g/L or 0.17 lb/gal)
  - 20% solution (wt/vol) of non-fat dry milk plus 0.1% polysorbate nonionic surfactant
Sanitation Measures

- Limit facility access to authorized personnel.
- Wear only clean clothing, and wash all clothing in hot water with soap prior to wearing again.
- If possible, use disposable protective clothing and gloves.
- Do not move protective clothing and tools from one area to another.
- Pull off gloves from the wrist upwards so that the glove turns inside out.
- Put all clothing into a hermetically sealed bag prior to exiting the area for washing or disposal.
- Put disposable clothing in the appropriate bin for immediate destruction.
- Use disinfectant mats at entrances for footwear and wheeled equipment and disinfect before entering and leaving areas.
- Disinfect tools before use on each plant (refer to the disinfection measures on page 2).
- Wash hands with soap or disinfectant before and after handling plants and putting on gloves.
Measures for Reducing Spread from Infected Areas

- Restrict movement:
  - Treat each infected area as a separate unit, and follow the sanitation measures listed on page 3.
  - Minimize movement between sites and move from a non-infected area to an infected area.

- Follow treatment measures on page 2:
  - Turn off irrigation water one day before plant removal to decrease the risk of sap transfer.
  - When removing plants and plant material, do not touch other plants or surfaces in greenhouses.

- Clean and disinfect susceptible materials and surfaces.
  - Follow disinfection measures on page 2 for anything that may have come into contact with infected plants and plant material including:
    - Tools,
    - Plant trays, which can also be treated by soaking in hot water at 90 °C (194 °F) for 5 minutes, or destroyed,
    - Drains, water storage areas, and irrigation system as the virus can survive in water; and
    - All agricultural machinery and equipment before moving between greenhouses.
  - Do not bring anything into an infected or suspected infected area that is not needed (jewelry, watches, phones, etc.) as everything will need to be disinfected before exiting the site.
  - Clean eye glasses with alcohol tissues after leaving infected areas.
  - Limit visitor access to a secure place with cleaning and disinfection facilities near the entrance to the greenhouse.
Follow cultural practices that reduce risk of ToBRFV spread:
  o Use naïve (young) bee hives for pollination. Avoid the use of bee hives in greenhouses if infection is found prior to flowering.
  o Keep records of seed and transplant sources for 3 years so they can be traced if necessary.
  o Eliminate potential ToBRFV reservoirs such as weeds. Follow treatment and disinfection measures on page 2 for any potential host material.
  o Scout plants for ToBRFV symptoms; take tissue samples and have diagnostic testing done on suspect plants.

Inform workers:
  o Institute hygiene and worker training programs that include sanitation of boots, clothing, hands, and tools in production areas.
  o Post informative materials on ToBRFV in production areas.

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