



Department
for Environment
Food & Rural Affairs

Pest specific plant health response plan:

Outbreaks of *Potato spindle tuber viroid* in tomato or
pepper crops



Figure 1. PSTVd infected tomato plant. © CSL via EPPO

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Executive summary

Background	
Regulation	GB Regulated Non-Quarantine pest
Key Hosts	Potato, tomato, pepper
Distribution	Africa, North America, South America, Asia, Europe, Oceania
Key pathways	Seed and ware potatoes, produce, plants for planting and seed
Industries at risk	Potato, tomato and pepper growers and packers
Symptoms	Stunting, chlorosis, distortion, purpling or reddening of leaves, reduction in fruiting and smaller spindle-shaped =potato tubers
Surveillance	
Demarcated zones (5.23)	Infected premises, neighbouring premises, other premises which staff/growers have visited or worked in, including those premises where machinery used was the same as that for the infected premises, stores, premises in which stock has been sent or received, and/or any other premises where there is a perceived risk.
Surveillance activities (5.21-5.22)	<ul style="list-style-type: none"> • Visual surveys will be carried out in the infested fields and at other propagators, producers and nurseries in the local vicinity • Sampling of symptomatic plants
Response	
Interceptions (5.1-5.4)	<ul style="list-style-type: none"> • Consignment should be destroyed or re-exported if dead larvae or feeding damage is seen. • Tracing exercises carried out where required • UKPHINs notification to be made.
Outbreaks (5.26-5.34)	<ul style="list-style-type: none"> • Destruction of infested plants and susceptible hosts within 20 m • Produce may move to retail or for processing if agreed by the IMT • Segregation of infested glasshouse compartments or gaps between rows • Post crop clean up, including decontamination of irrigation system
Key control measures	
Biological	N/A
Chemical	N/A
Cultural	Good hygiene practices, removal and destruction of infested plants
Declaration of eradication	
Eradication can be declared if no PSTVd is detected for a year following removal of the infested crop.	

Contents

Executive summary	3
1. Introduction and scope.....	5
2. Summary of threat	5
3. Risk Assessments.....	6
4. Actions to prevent outbreaks	6
5. Response.....	7
Official action to be taken following the suspicion or confirmation of PSTVd on imported plants, including seeds	7
Official action to be taken following the suspicion of PSTVd inland	7
Confirming a new outbreak	10
Criteria for determining an outbreak.....	11
Official Action to be taken following the confirmation of an outbreak	11
Review measures in the case of prolonged official action.....	14
6. Criteria for declaring eradication / change of policy	15
7. Evaluation and review of the contingency plan	15
8. Appendix.....	16
Data sheet for Potato spindle tuber viroid	16
9. References	7
10. Author and reviewers	21

1. Introduction and scope

- 1.1. This pest specific response plan has been prepared by the Defra Risk and Horizon Scanning Team. It describes how the Plant Health Service for England will respond if infection by *Potato spindle tuber viroid* (PSTVd) is discovered on potato (*Solanum tuberosum*).
- 1.2. The plant health authorities in Northern Ireland, Scotland, Wales and the Crown Dependencies have been consulted on this plan and will use it as the basis for the action they will take in the event of PSTVd being detected in their territories.
- 1.3. This document will be used in conjunction with the Defra Generic Contingency Plan for Plant Health in England (<https://planthealthportal.defra.gov.uk/assets/uploads/Generic-Contingency-Plan-for-Plant-Health-in-England-FINAL-2.pdf>), which gives details of the teams and organisations involved in pest response in England, and their responsibilities and governance. It also describes how these teams and organisations work together in the event of an outbreak of a plant health pest.
- 1.4. The aim of this response plan is to facilitate the containment and eradication of PSTVd.
- 1.5. This document can also be used as a basis for responding to outbreaks in potato of related pospiviroids. A datasheet containing background information on PSTVd is included in the appendix.

2. Summary of threat

- 2.1. *Potato spindle tuber viroid* (PSTVd) probably originates from Central America, but it was first identified in New Jersey, USA, in 1922 (Martin, 1922). It is now also present in South America, Europe, Africa, Asia and Oceania (EPPO, 2014). Within Europe, the viroid is widespread in Belarus and is present or occasionally present in Austria, Belgium, Croatia, Czech Republic, Georgia, Germany, Italy, Malta, Montenegro, Netherlands, Poland, Russia, Slovenia, Spain, Turkey and the Ukraine. The viroid is also reported as being under eradication in Greece, Hungary and Switzerland (EPPO, 2014).
- 2.2. The viroid causes growth reduction and other damaging symptoms in potato (*Solanum tuberosum*), tomato (*Solanum lycopersicum*) and pepper (*Capsicum annum*). Other *Solanum* spp. and some ornamental species in the family *Solanaceae* can also be infected, but the viroid is generally asymptomatic in these species. Yield losses of ~ 65% in potato have been recorded (Hunter and Rich,

1964), and potential yield losses of almost 100% may occur for tomato plants that are infected early (EFSA Panel on Plant Health, 2011).

- 2.3. In the UK, there have been two outbreaks of PSTVd, both on tomato, one in 2003 and one in 2011, and the viroid was eradicated on each occasion in the same year. There have also been several interceptions of the viroid in ornamental plants which have originated within the EU and multiple interceptions of the viroid on tomato and pepper seeds.

3. Risk Assessments

- 3.1. The viroid currently has an unmitigated and mitigated UK plant health risk register score of 75 and 45 respectively. Overall scores range from 1 (very low risk) to 125 (very high risk). These scores are reviewed as and when new information becomes available (<https://planthealthportal.defra.gov.uk/pests-and-diseases/uk-plant-health-risk-register/viewPestRisks.cfm?cslref=11980>).
- 3.2. The European Food Safety Authority (EFSA) published a scientific opinion on the risk of solanaceous pospiviroids for the EU territory in 2011.

4. Actions to prevent outbreaks

- 4.1. *Potato spindle tuber viroid* is a GB Regulated Non-Quarantine Pest (RNQP) when associated with ornamental plants for planting of pepper (*Capsicum annuum*), vegetable propagating and planting material (other than seeds) of pepper and tomato (*Solanum lycopersicum*), pepper and tomato seed, seed potatoes (*Solanum tuberosum*) and potato seeds as listed in Parts C, E, F, H and J of [Schedule 4 of The Plant Health \(Phytosanitary Conditions\) \(Amendment\) \(EU Exit\) Regulations 2020](#). There is a 0% threshold for findings of PSTVd on consignments of the listed commodities in GB, and measures to prevent their presence can be found in [Schedule 5](#) of the above regulations.
- 4.2. *Potato spindle tuber viroid* is an EU RNQP when associated with ornamental plants for planting of pepper (*Capsicum annuum*), vegetable propagating and planting material (other than seeds) of pepper and tomato (*Solanum lycopersicum*), pepper and tomato seed, seed potatoes (*Solanum tuberosum*) and potato seeds as listed in Parts D, F, G, I and K of Annex IV of Commission Implementing Regulation (EU) 2016/2031. There is a 0% threshold for findings of PSTVd on consignments of the listed commodities in EU.
- 4.3. *Potato spindle tuber viroid* is an A2 listed pest for the EPPO region and is therefore recommended for regulation by EPPO member countries.

- 4.4. The Plant Health Service (including the Animal and Plant Health Agency, Defra and Fera Science Ltd.) should be aware of the measures described in this plan and be trained in responding to an outbreak of PSTVd in potato crops. It is important that capabilities in detection, diagnosis and risk management are available.

5. Response

Official action to be taken following the suspicion or confirmation of PSTVd on imported plants, including seeds

Holding consignments at ports of entry

- 5.1. If PSTVd is suspected by the Plant Health and Seeds Inspectorate (PHSI) to be on a consignment moving in trade, the PHSI should hold the consignment until a diagnosis is made. Samples should be sent in by the PHSI to Plant Clinic, Fera Science Ltd., York Biotech Campus, Sand Hutton, York, YO41 1LZ (01904 462000), in a sealed bag or contained, within at least two other layers of containment which are not liable to be crushed during transit.
- 5.2. If PSTVd is confirmed, the PHSI should advise the client of the action that needs to be taken by way of an official notice. The consignment should be destroyed by either incineration or deep burial. This is most likely to occur for seeds which are tested at the point of entry.
- 5.3. If PSTVd is confirmed, a UKPHINS (UK Plant Health Interception Notification System) notification should be made. UKPHINS is the IT system for recording findings and non-compliance in order to maintain records and notify other National Plant Protection Organisations (NPPO) of plant health issues.
- 5.4. If all or part of the consignment has been distributed to other premises prior to diagnosis, trace forward inspections should take place upon suspicion or confirmation of PSTVd.

Official action to be taken following the suspicion of PSTVd inland

- 5.5. Suspected outbreaks will be assessed on a case by case basis. An Outbreak Triage Group (OTG), chaired by the Chief Plant Health Officer (CPHO) or their deputy and including specialists from APHA, Defra and other organisations, should be set up to assess the risk and decide on a suitable response. Where appropriate, the OTG will also decide who will be the control authority, and the control authority will then

nominate an incident commander. An Incident Management Team (IMT) meeting, chaired by the Incident Commander, will subsequently convene to produce an Incident Action Plan (IAP). See the Defra Generic Contingency Plan for Plant Health in England for full details.

- 5.6. The OTG will determine the alert status, which will consider the specific nature of the outbreak. These alert levels, in order of increasing severity, are white, black, amber and red (more detail on these levels can be found in table 2 of the *Defra Generic Contingency Plan for Plant Health in England*). Following suspicion of PSTVd in tomato and pepper (under protection), a black alert status should generally be instigated by the PHSI. The black alert status is used for a significant plant disease that has the potential for geographic spread.
- 5.7. If PSTVd is suspected (or confirmed) in an allotment or garden then this will be dealt with on a case by case basis.

Restrictions on movement of material, equipment and machinery to and from the place of production

- 5.8. *Potato spindle tuber viroid* can be transmitted mechanically, on gloves and hands (van Brunsschot *et al.*, 2014), on machinery (Merriam and Bonde, 1954) and on tools (Verhoeven *et al.*, 2010b). The viroid can also remain infective on the outside of plants for at least 8 weeks (Mumford *et al.*, 2004b). Movement of material, equipment and machinery between infected and non-infected areas should therefore be restricted. However, if movement is necessary, the material, equipment and machinery should be thoroughly cleaned and disinfected (see 5.24).

General biosecurity advice and advisory measures for growers

- 5.9. Hygiene best practice, primarily relating to places of production or propagation, should be followed (EFSA Panel on Plant Health, 2011). This includes:
 - Training staff to identify symptoms of PSTVd, and to follow best practice procedures.
 - Prohibiting the consumption of fruit of susceptible species on the premises. Handling infected fruit could contaminate the hands, which in turn could infect the crop.
 - Prohibiting the sorting/packing of fruit produced from other companies/locations. If the fruit is infected, machinery, equipment and people could become contaminated and infect other crops on the premises.
 - Using clothes (including overshoes), which will either be destroyed (via incineration or deep burial) or washed following work on a particular lot. This prevents spread between lots.

- Using disposable gloves that will be destroyed (via incineration or deep burial) following work on a particular crop, between different areas within a crop or between plants (reducing spread).
- Washing hands with soap before and after entering a new glasshouse or compartment (reducing spread).
- Restricting the use of equipment, particularly knives, to one location, to prevent the viroid spreading to other locations (via mechanical transmission).
- Chemical disinfection of knives and pruning instruments (between crops, areas within a crop or plants to reduce spread) (see 5.24).
- Cleaning and disinfection of machinery between crops. As with handheld equipment, machinery is another means of mechanical transmission. Disinfection and cleaning of machinery with high water pressure, steam cleaners or other methods may therefore reduce spread. Records of this should be maintained.
- Maintaining the working direction. If human-assisted spread of a pathogen occurs, it will occur in the direction that the human is working. Working in the same direction reduces the extent of the spread and allows measures to be carried out in a more concentrated way.
- Restricting access to the working area. The fewer people entering a particular lot, the less chance there is that PSTVd will be introduced. Wherever possible, employees should work in the same areas or number of rows each day rather than swapping around work areas.

5.10. Volunteer plants and weeds, particularly perennials, act as reservoirs for PSTVd (EFSA Panel on Plant Health, 2011). Controlling these plants reduces the chance of the crop becoming infected. Volunteer plants and weeds arising in potato cultivations can be controlled mechanically (e.g. by hoeing, roguing, flame weeding), chemically (e.g. using herbicides) and by cultural practices (e.g. lifting of potatoes). It is important there is no 'carry over' into the next crop by self-sown seedlings arising from seed of squashed fruit from the previous season. Any disposal required should be as in point 5.35-36.

5.11. Transmission of PSTVd by aphids has occasionally been reported in potato within and between species but transmission is at a very low level. Research suggests that higher levels of transmission have been observed when the viroid has been acquired from potato plants co-infected with Potato leafroll virus (PLRV) (e.g. for *Myzus persicae*, Syller *et al.*, 1997). Controlling aphids in crops, particularly those crops that are also hosts of PLRV, is therefore recommended.

Preliminary trace forward / trace backward

5.12. Likely pathways include true seed from non-EU countries and plants for planting of tomato and pepper that have been grown from infected seed in EU countries. Seed that is labelled as 'EU origin' may have been produced in third countries and only cleaned and re-packed in an EU country, so the seed company will need to be

contacted to confirm this. If an infested consignment is considered as being the source of the suspect outbreak, investigations regarding the origins of infested consignments will be undertaken to locate other related and therefore potentially infested consignments moving to and from the site. If applicable the relevant NPPO should be contacted. This process is particularly important for propagation or seed stock.

Confirming a new outbreak

How to survey to determine whether there is an outbreak

5.13. Information to be gathered on the suspicion of PSTVd by the PHSI, in accordance with ISPM 6; guidelines for surveillance (<https://www.ippc.int/en/publications/615/>):

- The origin of the host plant(s) and relevant plant and seed lot numbers etc.
- Details of other premises or destinations where the plants have been grown or sent, where the viroid may be present.
- The layout of the premises and surrounding area, including a map of the cultivations/buildings, at risk growers, any other host plants, including susceptible ornamentals etc.
- Details of the host plant: the species, variety, growth stage and any other relevant information. For tomato plants, which are often grafted onto rootstocks, details of the rootstocks are also necessary.
- Description of the surrounding habitat and climate.
- Level of infection, including a description of symptoms (could take photos).
- The date and time the sample was taken, how it was identified and by whom.
- Current controls in place e.g. chemical treatments (These cannot be used against the internal viroid infection, but may be used for insect control. Chemicals may also distort the appearance of symptoms, reducing the effectiveness of visual survey).
- Details on the movement of people, equipment, machinery etc. to and from the infected area.
- Cultural and working practices.
- History of PSTVd on the site and nearby, if any.
- The presence of aphids and PLRV.

5.14. Further to information gathering, samples of other symptomatic host plants should be taken to confirm the extent of infection e.g. in surrounding lots. This initial survey will be used to determine if it is an isolated case or an established outbreak.

5.15. Finance for the surveys will depend on the individual circumstances of the outbreak, and will be subject to discussion.

Sampling

- 5.16. Following the identification of a suspect plant, symptomatic parts of the plant (e.g. leaves, fruit and stems) should be sent to Fera Science Ltd. as in 5.1. Plants should be handled with gloves. Gloves should be destroyed (via incineration or deep burial) following use. Hygiene best practice should be followed while sampling. Each sample should be labelled with full details of sample number, location, variety etc.

Diagnostic procedures

- 5.17. The principal means of detecting pospiviroids is through reverse transcription-polymerase chain reaction (RT-PCR) (Shamloul *et al.*, 1997; Bostan *et al.*, 2004; Verhoeven *et al.*, 2004; Verhoeven *et al.*, 2011). At Fera, TaqMan is used. This method is generally non-specific and will not allow identification down to species level (e.g. Singh *et al.*, 1999; Verhoeven *et al.*, 2011). Real-time RT-PCR is also often run in tandem, which narrows down the viroid species to either PSTVd or TCDVd (Boonham *et al.*, 2004). Identification to species level and separation from similar viroids such as TCDVd requires sequencing of the RT-PCR products and BLAST analysis (Boonham *et al.*, 2005; Verhoeven, 2010a). This is in accordance with ISPM 8 (<https://www.ippc.int/en/publications/612/>) and ISPM 27 Annex 7 (<https://www.ippc.int/en/publications/593/>).

Criteria for determining an outbreak

- 5.18. If PSTVd is detected at a location other than at a port or confined to a particular consignment with no risk of spread (e.g., tomato seed), then an outbreak should be declared.

Official Action to be taken following the confirmation of an outbreak

- 5.19. The scale of the outbreak will determine the size and nature of the management team and action.

Communication

- 5.20. The IMT will assess the risks and communicate details to the IPPC, EU and EPPO, in accordance with ISPM 17: pest reporting (<https://www.ippc.int/en/publications/606/>), as well as within Government to Ministers, senior officials, devolved administrations and other government departments and agencies (e.g., the Environment Agency) on a regular basis as appropriate; and to other relevant stakeholders.

Surveillance

- 5.21. Host plants on the affected premises should be visually inspected at a surveying rate determined by the IMT. This should focus primarily on tomato and pepper. Symptomatic plants, including host species other than tomato and pepper, should also be tested. These may include host plants adjacent to the affected premises, such as weeds, if present.
- 5.22. Propagators, nurseries and producers within the nearby surrounding area should be surveyed. Other premises which staff/growers have visited or worked in and any premises where there is a perceived risk should also be inspected.

Demarcated zones

- 5.23. The premises and immediate vicinity (neighbouring host plants), other premises in which staff/growers have visited or worked in, premises in which stock has been sent or received, and/or any other premises where there is a perceived risk, should be demarcated for surveillance.

Decontamination procedures

- 5.24. Thorough cleaning and application of disinfectants should be used for all non-disposable material, equipment and machinery. Virkon S (2%, containing potassium peroxymonosulfate) and sodium hypochlorite (e.g. 10% Clorox regular bleach) are recommended disinfectants (Li *et al.*, 2015). Any waste (plant or other potentially infected material) should be removed and destroyed (via deep burial, incineration or other appropriate methods prescribed in 5.35).

Tracing forwards / backwards

- 5.25. Once other sites that are potentially infected by PSTVd have been identified, these should be inspected as per surveillance highlighted in paragraphs 5.21 - 5.22. Information, which is aimed at raising awareness of the disease and its symptoms, should be sent to affected and at risk growers. A pest factsheet can be found on the plant health portal - <https://planthealthportal.defra.gov.uk/plant-health-api/api/pests/11980/notices/6722/documents/4255/document>.

Pest Management procedures

- 5.26. Host plants should not be moved off site, with exception to fruit that may be moved off site for retail if agreed by the IMT.
- 5.27. There are no chemical or biological methods for controlling PSTVd. Therefore, the only effective method of eradication is destruction. All infected plants, at risk plants along the row that are within 20 m of the infected plants, plants an equivalent

distance in rows either side of the infected row, and volunteer plants, should be destroyed; and if there are several outbreaks within the same crop, it is advised that the whole crop is destroyed. However, see the advice in 5.29 concerning large tomato and pepper plants. In addition, if an outbreak is found within a breeding or propagation lot, all plants in the lot should be destroyed, even if the outbreak is only found in a single spot, because of the potential for the viroid to be present in or spread throughout the whole lot.

- 5.28. To reduce spread before or during the process of destruction, glasshouses containing infected plants can be segregated by introducing compartments or, if the plants are small, moving them to create a gap between stocks (e.g. 10 m).
- 5.29. Removal of large plants (especially mature tomatoes and peppers in a glasshouse) is not advised as this is likely to increase the risk of spread through physical contact between infected and uninfected plants, debris and machinery, personnel etc. If measures can be put in place to contain the viroid by leaving plants in situ until the end of the season (crop cycle) when the whole crop is removed, it is likely to be the preferred option.
- 5.30. A limited number of employees should work in the outbreak area, and the work should be completed at the end of the day to avoid spreading the viroid to other areas. The working direction described in 5.9 should also be used to delineate areas. For example, the three rows in the working direction and one row in the opposite direction can be marked off and treated separately at the end of the day.
- 5.31. If harvesting of fruit from the infected crop continues, fruit should be packed on site and go direct to retail or wholesale and not be repacked at another production site unless hygiene measures can be put in place to ensure cross contamination is avoided.
- If agreed by the IMT, produce may be moved to other production sites for packing providing there are suitable hygiene measures in place to prevent cross contamination of growing crops.
 - It should be ensured that there is no risk of spread to other production sites from the reuse of packaging used for transport of infested produce under a statutory plant health notice.
 - A notice will also be issued to sites where infested produce is sent prior to being moved for retail/wholesale.
- 5.32. On most tomato and pepper production sites there will be a crop break. During this break, all plants should be removed and appropriately disposed of by burning or deep burial (with other methods considered on a case by case basis). On sites which practice all year round cropping, successive crops are planted next to each other to reduce the period between marketable crops. Therefore, infection within one year's crop would be very likely to spread to the next years' crop as a result. Where continuous cropping is practiced, a crop break will be needed to eradicate the viroid. To reduce the risk of further spread, additional host crops should not be planted in

the same facility whilst infected plants are present. Host crops should only be planted in nearby glasshouses, if suitable measures to prevent spread can be put in place.

- 5.33. Once the infected crop has been removed, all remaining material, e.g., string, plastic flooring and growing media, should be destroyed and the facility thoroughly cleaned with water and detergent to remove any remaining plant material and finally disinfected with a suitable disinfectant.
- 5.34. Water is also a potential route of transmission (Mehle *et al.*, 2014), but evidence to support this under normal growing conditions is limited. As a precaution, the irrigation system should be decontaminated and cleaned out at the end of the season. Water for hydroponic and irrigation systems should subsequently come from sources free from the viroid, and, if possible, water should not be mixed between infected and non-infected lots.

Disposal plan

- 5.35. The primary means of disposing of infected material and plants is through incineration (licensed) and deep burial. Deep burial may be done at an approved landfill site, or on the site or nearby farm, if practical and in agreement with the local Environment Agency. Incineration must comply with appropriate waste management regulations, Environment Agency in England, Scottish Environment Protection Agency and Natural Resources Wales. If the material has to be moved off the premises, it should be contained within at least two sealed layers, if possible (e.g. small plant within two plastic bags).
- 5.36. Aside from incineration and deep burial, other viable methods of destruction may include anaerobic digestion and recycling (e.g., of rockwool slabs for non-horticultural use). However, these and any other methods should be agreed by the incident management team.

Review measures in the case of prolonged official action

- 5.37. Monitoring of affected premises and demarcated areas should take place. In the following year, this should take place monthly throughout the growing season for glasshouse tomato and pepper. Plants should be visually inspected and any plants showing suspect symptoms should be tested. Any volunteer plants should be removed and destroyed by burning or deep burial.
- 5.38. The EPPO protocol states that if continuing official action is required within the demarcated area over a prolonged period, a review of eradication and containment measures should be undertaken regularly to determine the success and cost-

effectiveness of measures in the longer term. This review will involve consultation with stakeholders and should include:

- Evaluation of the effectiveness of current measures
- Evaluation of the economic impact and cost-effectiveness of continuing existing measures
- Consideration of further measures to strengthen containment and eradication actions
- Consideration of statutory obligations and impact on import and export procedures
- Consideration of alternative approaches, including pursuing measures to contain the pest rather than eradication or even the cessation of statutory action.

In circumstances where it is considered that the pest cannot be eradicated or contained and official action is no longer considered appropriate, stakeholders should be consulted and a timetable and mechanism for the removal of official measures, and for the dissemination of pest management information, should be agreed by the LGD.

6. Criteria for declaring eradication / change of policy

- 6.1. *Potato spindle tuber viroid* can be declared eradicated (by the Chief Plant Health Officer Unit) in tomato and pepper if it has not been found for a year (or for a single cycle of the crop) after the infected crop was removed.

7. Evaluation and review of the contingency plan

- 7.1. This pest specific contingency plan should be reviewed regularly to consider changes in legislation, control procedures, sampling and diagnosis methods, and any other relevant amendments.
- 7.2. Lessons should be identified during and after any PSTVd or non-PSTVd outbreak, including what went well and what did not. These should be included in any review of the contingency plan leading to continuous improvement of the plan and response to outbreaks.

8. Appendix

Data sheet for Potato spindle tuber viroid

Identity

PREFERRED SCIENTIFIC NAME	AUTHOR (taxonomic authority)
<i>Potato spindle tuber viroid</i>	Diener (1971)

SUPERKINGDOM: Viroids

FAMILY: Pospiviroidae

GENUS: Pospiviroid

NON- PREFERRED SCIENTIFIC NAME (EPPO, 2014)	AUTHOR (taxonomic authority)
<i>Potato gothic virus</i>	-
<i>Potato spindle tuber pospiviroid</i>	-
<i>Potato spindle tuber virus</i>	-
<i>Tomato bunchy top viroid</i>	-

INTERNATIONALLY USED COMMON NAME(S) AND INTERNATIONAL LANGUAGE (EPPO, 2014):

Spindelknollenkrnakheit (German)

Bunchy top of tomato (English)

Spindle tuber of potato (English)

Notes on taxonomy and nomenclature

There are two viroid families; Avsunviroidae and Pospiviroidae. They are distinguished based on the presence or absence of a Central Conserved Region (CCR) and hammerhead ribozymes. The family Pospiviroidae has a CCR and does not form hammerhead ribozymes. This family is composed of five genera, including the genus Pospiviroid. *Potato spindle tuber viroid* (PSTVd) is the type species of the Pospiviroid genus (CABI, 2015; Flores *et al.*, 2009).

Species are discriminated from one another based on their level of sequence similarity across the whole genome; if their sequences differ by more than 10%, they are classified as separate species (Flores *et al.*, 2005). However, some genetically similar viroids are also separated because of differences in host range and symptoms. This is the case for PSTVd and *Tomato chlorotic dwarf viroid*.

Biology and ecology

Once the viroid has been transmitted into a host plant cell, it replicates within the nuclei via a rolling circle mechanism (Flores *et al.*, 2009).

Hosts/crops affected

The main host of PSTVd is considered to be potato (*Solanum tuberosum*), primarily because of the severity of symptoms that are seen on the plant following infection. Other hosts which suffer symptoms are *Solanum lycopersicum* (tomato), *Capsicum annuum* (pepper) and *Physalis peruviana* (Cape gooseberry) (Mackie *et al.*, 2002; Lebas *et al.*, 2005; Ward *et al.*, 2010). Symptomless infections have also been reported from , *Brugmansia* spp., *Chrysanthemum* sp., *Calibrachoa* sp., *Cestrum* spp., *Dahlia* sp., *Datura* sp. *Ipomoea batatas* (sweet potato), *Lycianthes rantonnei*, *Persea americana* (avocado), *Petunia* sp., *Solanum pseudocapsicum*, *Streptosolen jamesonii*, *Solanum jasminoides*, *Solanum muricatum* and wild *Solanum* spp. (Salazar, 1989; Owens *et al.*, 1992; Querci *et al.*, 1995; Behjatnia *et al.*, 1996; Di Serio, 2007; Verhoeven *et al.*, 2008a, b, 2009, 2010; Lemmetty *et al.*, 2011; Luigi *et al.*, 2011; Mertelik *et al.*, 2010; Verhoeven, 2010b; Tsushima *et al.*, 2011). The experimental host range of the viroid is even wider, numbering over 130 species, including several solanaceous plants (e.g. *Solanum melongena* (aubergine)) and species from Amaranthaceae, Asteraceae, Boraginaceae, Campanulaceae, Caryophyllaceae, Compositae, Convolvulaceae, Dipsacaceae, Orobanchaceae, Sapindaceae, Scrophulariaceae and Valerianaceae (Singh, 1973; Matoušek *et al.*, 2007; Vachev *et al.*, 2010; Matsushita and Tsuda, 2015).

Plant stages affected

Potato spindle tuber viroid affects the flowering stage, fruiting stage and the vegetative growing stage.

Plant parts affected

Potato spindle tuber viroid affects the leaves, roots, fruits and tubers, as well as the size of the plant.

Symptoms/signs – description

Potato

Growth is often reduced, but impacts range from very mild (barely noticeable) to severe (CABI, 2015). There is an accumulation of pigment at the top of stems, which is generally associated with the rolling of terminal leaflets. If viewed from above, an infected potato plant is considered to exhibit clockwise phyllotaxy (= arrangement of leaves on the plant stem) (EPPO data sheet). Vines may be smaller, more spindly and upright. Leaves may also be smaller, as well as darker and more crinkled (CABI, 2015). Axillary buds

sometimes proliferate to give the impression of Witches' Broom (EPPO data sheet). Further down the plant, tubers lose their shape, some becoming thinner and smaller, while others become bigger ('giant hill') (Gilbert, 1925; CABI, 2015; EPPO data sheet). The tubers also exhibit a cracked appearance (CABI, 2015). The eyes of the tubers are sometimes more pronounced, increase in number and may be borne on 'knob-like protuberances' (Martin, 1922, 1924; CABI, 2015). Sprouting might also be slower than normal (EPPO data sheet).

The type and severity of symptoms vary depending on the strain of PSTVd and the cultivar of potato (Singh *et al.*, 1971; Pfannenstiel and Slack, 1980; Kowalska-Noordam *et al.*, 1987; Nakahara *et al.*, 1997). For example, Macleod (1927) found symptoms to be more obvious on Irish Cobbler than on Oreen Mountain Potatoes. Environmental conditions have also been shown to impact on symptoms, with higher soil moisture and temperature resulting in more serious damage of tubers (Goss, 1930).

Tomato

The first signs of PSTVd are growth reduction, epinasty (drooping of the leaf, caused by greater growth on the upper surface), chlorosis and crinkling at the top of the plant (Fig. 2; CABI, 2015; EPPO data sheet). This is followed by more severe chlorosis lower down the plant, which eventually results in reddening and purpling, and/or necrosis, and the leaves becoming brittle (Fig. 3; CABI, 2015). The growth reduction at the top of the plant may also lead to stunting of the whole plant (CABI, 2015). This is coupled with the cessation of flower and fruit initiation (CABI, 2015). In the worst case, stunting is followed by the death of the plant.

Pepper

Outside of the laboratory, the only symptoms observed on pepper have been the distortion of the leaf margins at the top of the plant (Lebas *et al.*, 2005). Artificial inoculation of the viroid has also resulted in a reduction of fruit size (Verhoeven *et al.*, 2009a).

Other fruit and vegetable crops, and ornamentals

Infection outside of potato, tomato and pepper is generally asymptomatic, though there is a record of symptom expression in Cape gooseberry plants (Ward *et al.*, 2010). Symptoms have also been displayed in aubergine and *Petunia x hybrida* following experimental inoculation (Matsushita and Tsuda, 2015).

Morphology

Potato spindle tuber viroid is a small, circular, single stranded RNA (Gross *et al.*, 1978). Depending on the strain, the number of nucleotides can total between 356 and 363 (Puchta *et al.*, 1990; Lakshman and Tavantzis, 1993; Behjatnia *et al.*, 1996; Verhoeven *et al.*, 2010a).



Figure 2. Epinasty, chlorosis and leaf crinkling of tomato plant.
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Figure 3. Purpling of tomato leaves.
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Similarities to other species/diseases/plant damages

The host range and symptom expression of other pospiviroids is similar to PSTVd (CABI, 2015). At least seven pospiviroids outside of PSTVd have been recorded naturally infecting tomato, and *Pepper chat fruit viroid* (PCFVd) has been recorded naturally infecting pepper (EFSA Panel on Plant Health, 2011). However, only PSTVd has been confirmed to infect potato in nature. All other pospiviroid infections of potato have been the result of experimental transmission (EFSA Panel on Plant Health, 2011).

Detection and inspection methods

Visual inspection of potato and tomato allows for the detection of pospiviroids if symptoms are present. However, because symptoms are similar between pospiviroids, it is difficult to distinguish PSTVd from other species. Depending on the severity of the strain, the environmental conditions and the host, symptoms may also not be evident. Laboratory tests are therefore required.

The principal means of detecting pospiviroids is through reverse transcription-polymerase chain reaction (RT-PCR) (Shamloul *et al.*, 1997; Bostan *et al.*, 2004; Verhoeven *et al.*,

2011). This method is generally non-specific and will not allow identification down to species level (e.g. Singh *et al.*, 1999; Verhoeven *et al.*, 2011). Identification to species level requires sequencing of the RT-PCR products and BLAST analysis (Boonham *et al.*, 2005; Verhoeven, 2010a).

Other methods that allow for the detection of pospiviroids include the use of indicator plants (Raymer *et al.*, 1964; Fernow *et al.*, 1969; Singh, 1984; Grasmick and Slack, 1987), gel electrophoresis (Morris and Wright, 1975; Schumann *et al.*, 1978; Schumacher *et al.*, 1986), nucleic acid hybridization (Owens and Diener 1981; Salazar *et al.*, 1983, 1988a, Lakshman *et al.*, 1986; Roy *et al.*, 1989; Candresse *et al.*, 1990; Podleckis *et al.*, 1993; Singh *et al.*, 1994; Khan *et al.*, 2009; Monger and Jeffries, 2015), real-time RT-PCR (Boonham *et al.*, 2004; Roenhorst *et al.*, 2005), reverse transcription loop-mediated isothermal amplification (RT-LAMP) (Tsutsumi *et al.*, 2010), real-time RT-LAMP (Lenarcic *et al.*, 2012) and macro/microarrays (Agindotan and Perry, 2008; Tiberini and Barba, 2012).

Distribution

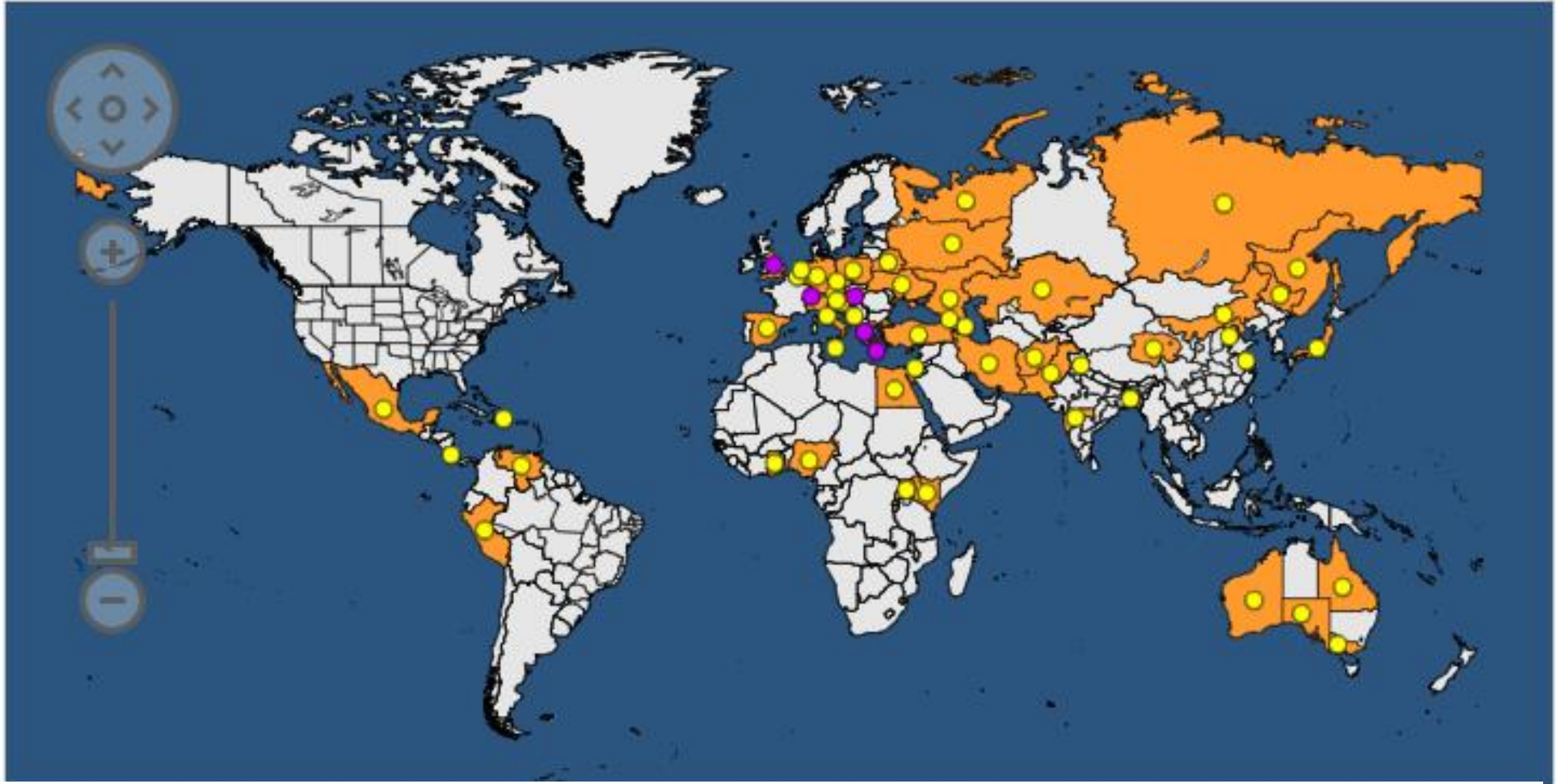


Figure 2. Distribution of PSTVd (yellow = present, purple = transient) (Source EPPO Global database). Up to date distribution data can be found using the link below.

<https://gd.eppo.int/taxon/PSTVD0/distribution>

History of introduction and spread

It has been suggested that PSTVd, along with *Mexican papita viroid* and *Tomato plancho macho viroid*, descends from a common ancestor originating in Mexico, on a wild *Solanum cardiophyllum* plant (Hoop *et al.*, 2008). However, spindle tuber disease was first identified in New Jersey, USA, in 1922 (Martin, 1922), and soon after in Maine (Martin 1922; Schulz and Folsom 1923). The viroid has since spread to a number of countries across various continents (Fig. 2). Reports of the viroid in potato fields have come from the USA, Canada, China, Russia and Turkey (Singh *et al.*, 1970, 1991, 1993b; Tien, 1985; He *et al.*, 1987; Güner *et al.*, 2012). The USA and Canada have since eradicated PSTVd (Sun *et al.*, 2004; De Boer *et al.*, 2005). The viroid has also been recorded in potato collections and breeding material in the UK, Australia, Argentina, Peru, the Netherlands, Venezuela and Brazil (Cammack and Richardson, 1963; Scottish Plant Breeding Station, 1976; Schwinghamer *et al.*, 1983; Cartwright, 1984; Fernandez-Valiela, 1965; Singh, 1983; Netherlands NPPO, 2014), but has been eradicated in the UK and Argentina. In tomato, reports of PSTVd have come from Australia, Belgium, Italy, Japan, New Zealand, the Netherlands, UK and the USA (Puchta *et al.*, 1990; Elliott *et al.*, 2001; Mackie *et al.*, 2002; Hailstones *et al.*, 2003; Mumford *et al.*, 2004a; Verhoeven *et al.*, 2004, 2007; Matshushita *et al.*, 2008; Navarro *et al.*, 2009; Ling and Sfetcu, 2010). Further, there have been a number of reports of PSTVd in other crops, such as ornamentals and peppers, in other countries (e.g. EFSA Panel on Plant Health, 2011; Lebas *et al.*, 2005).

Phytosanitary status

Potato spindle tuber viroid is a GB Regulated Non-Quarantine Pest (RNQP) when associated with ornamental plants for planting of pepper (*Capsicum annuum*), vegetable propagating and planting material (other than seeds) of pepper and tomato (*Solanum lycopersicum*), pepper and tomato seed, seed potatoes (*Solanum tuberosum*) and potato seeds as listed in Parts C, E, F, H and J of Schedule 4 of The Plant Health (Phytosanitary Conditions) (Amendment) (EU Exit) Regulations 2020. There is a 0% threshold for findings of PSTVd on consignments of the listed commodities in GB.

It is also present on several other phytosanitary lists.

Table 1. Global phytosanitary categorisation of PSTVd.

Country/NPPO/RPPO	List	Year of addition
AFRICA		
East Africa	A1 list	2001
Egypt	A1 list	2018
Morocco	Quarantine pest	2018

Country/NPPO/RPPO	List	Year of addition
Tunisia	Quarantine pest	2012
AMERICA		
Argentina	A1 list	2019
Brazil	A1 list	2018
Canada	Quarantine pest	2019
Chile	A1 list	2019
Mexico	Quarantine pest	2018
USA	Quarantine pest	1989
Uruguay	A1 list	1995
ASIA		
Bahrain	A1 list	2003
Israel	Quarantine pest	2009
Jordan	A1 list	2013
Kazakhstan	A1 list	2017
EUROPE		
GB	RNQP	2020
Georgia	A1 list	2018
Moldova	Quarantine pest	2017
Norway	Quarantine pest	2012
Russia	A2 list	2014
Turkey	A2 list	2016
Ukraine	RNQP	2019
RPPO		
COSAVE	A2 list	2018
EAEU	A2 list	2016
EPPO	A2 list	1975
EU	RNQP	2019

Means of movement and dispersal

There is a high transmission rate of PSTVd from mother plants to their vegetatively propagated progeny (Owens and Verhoeven, 2009). This vegetative propagation material can be moved over long distances in trade, and is thought to be a major source of spread in potato and ornamental plants, especially in the absence of symptoms (Singh *et al.*, 1993b; Di Serio, 2007; Owens *et al.*, 2009).

A further form of human assisted transmission is mechanical spread. The spread of PSTVd has been shown between plants of the same and different species, via foliage and tuber contact (Bonde and Merriam, 1951; Merriam and Bonde, 1954), gloves and hands (Siegener *et al.*, 2008; Verhoeven *et al.*, 2010b; Fujiwara *et al.*, 2013; van Brunschot *et al.*, 2014 [fruit sap]), machinery (Merriam and Bonde, 1954; Manzer and Merriam, 1961), and tools (Verhoeven *et al.*, 2010b; Fujiwara *et al.*, 2013). PSTVd remains infective for hours outside of plants, as is shown on hands and gloves (Verhoeven *et al.*, 2010b; van Brunschot *et al.*, 2014). In addition, transmission efficiency varies depending on the recipient host and the source of inoculum (Verhoeven *et al.*, 2010b). For example, PSTVd was more readily transmitted to tomato and potato from *Solanum jasminoides* than from *Brugmansia suaveolens*. Verhoeven *et al.* (2010b) has also shown that temperature is a factor, with 25°C favouring transmission of PSTVd and 15°C being inhibitory.

Transmission by seed and pollen results in spread within a species. This method of transmission for PSTVd has been reported for potato (Hunter *et al.*, 1969; Singh, 1970; Singh *et al.*, 1992), tomato (Benson and Singh, 1964; Singh, 1970; Kryczynski *et al.*, 1988; van Brunschot *et al.*, 2014), and *Scopolia sinensis*, a wild solanaceous plant (Singh and Finnie, 1973), as well as experimentally by Kryczynski *et al.* (1992) and van Brunschot *et al.* (2014). The rates of transmission in potato vary between collections (Fernow *et al.*, 1970; Singh *et al.*, 1993b). EUPHRESCO (2011) have also shown that the viroid accumulates within the tissues of tomato seeds, which might leave it protected (either fully or partially) from disinfection techniques.

Transmission by insect vectors also allows for spread within and between species. Early experiments by Kennedy *et al.* (1962) and Smith (1972) showed that the aphids *Macrosiphum euphorbiae* and *Myzus persicae* transmitted PSTVd in potato. However, a later study by Schumann *et al.* (1980) did not corroborate these findings. Further, in a study by De Bokx and Piron (1981), PSTVd was not transmitted by *M. persicae* or *A. solani*, and was only transmitted by *M. euphorbiae* to tomato with very low efficiency. More recent research has shown that PSTVd is readily transmitted by *M. persicae* in the presence of *Potato leafroll virus* (PLRV) in potato (Salazar *et al.*, 1995). The presence of the virus might even be a necessity for *M. persicae*, with PSTVd failing to be transmitted in the absence of the virus in both potato and tomato (Querci *et al.*, 1997; Singh and Kurz, 1997; Syller *et al.*, 1997 [from *P. floridana*]). Syller *et al.* (1997) also showed that PSTVd was encapsidated by (enclosed within the protein shell of) PLRV, rather than simply being adsorbed onto the outside, and that this might explain the successful aphid transmission.

Although bumblebees have shown transmission of *Tomato apical stunt viroid* (TASVd) and *Tomato chlorotic dwarf viroid* (TCDVd) (Antignus *et al.*, 2007; Matsuura *et al.*, 2010), a study by Nielsen *et al.* (2012) did not demonstrate transmission of PSTVd in this way. Nielsen *et al.* (2012) also did not show transmission of the viroid in adult thrips, though encapsidation was not explored, and nor was feeding by nymphs.

Mehle *et al.* (2014) has recently shown that PSTVd can remain infective within water for up to 7 weeks, and that infected water can lead to the infection of tomato roots and later the infection of the green parts of the plant. Potato tubers developed from plants grown in PSTVd infected water were also shown to be sources of the viroid. In hydroponic systems, where water is recycled, tomato and pepper plants are grown for several months, and sometimes a year. There is therefore time for the viroid to accumulate in or on roots. Although, it should be noted that infection efficiency was low, and that if infection did occur, movement into the rest of the plant was delayed and unevenly distributed.

Control

Chemical and biological control

There are currently no effective chemical or biological control options for PSTVd itself (CABI, 2015). However, its aphid vectors can be controlled through traditional means (EFSA Panel on Plant Health, 2011).

Resistant crop cultivars

There are currently no naturally occurring plant cultivars that offer full resistance to PSTVd. *Solanum acaule* offers partial resistance and is impervious to mechanical inoculation by PSTVd, but is vulnerable to infection when cDNAs containing the viroid are used in agroinfection (Salazar *et al.*, 1988b). Partial resistance has also been reported for other plants, including *Solanum guerreroens* and *Solanum berthaultii* (Harris *et al.*, 1979; Pfannenstiel and Slack, 1980; Singh, 1985; Palukaitis, 2012). Experimentally, resistance to PSTVd has been shown by Yang *et al.* (1997), Sano *et al.* (1997) and Schwind *et al.* (2009).

Cultural controls and sanitary methods during cultivation

Hygiene best practice

Principles of hygiene best practice for pospiviroids, including PSTVd, are outlined in the EFSA Panel on Plant Health (2011) and are briefly described in the contingency plan (Precautionary measures section).

Cleaning and disinfectants

Thorough cleaning of a glasshouse using a steam cleaner, and a scrub brush for less easily accessible structures, together with detergent is advised. An acid treatment for watering tubes and drippers has also been suggested by Owens and Verhoeven (2009).

Both Virkon and Virkon S (Li *et al.*, 2015; Olivier *et al.*, 2015), and sodium hypochlorite (e.g. Clorox regular bleach, Singh *et al.*, 1989; Roenhorst *et al.*, 2005; Li *et al.*, 2015) have been shown to be particularly effective as disinfectants of PSTVd. Nonfat dry milk, Lysol all-purpose cleaner, Viroid, Hyprelva SL and Jet S have also been shown to have a marked effect on PSTVd (Li *et al.*, 2015; Olivier *et al.*, 2015). Although Menno Florades and MENNO clean are mentioned in EPPO (2011) as suitable disinfectants for PSTVd, they were not demonstrated to be that effective by Li *et al.* (2015) or Olivier *et al.* (2015), respectively. It should be noted that disinfectants were less effective when applied to dried sap droplets infected with PSTVd (Olivier *et al.*, 2015). This was attributed to thick halos of sap forming at the periphery of the droplets, allowing the viroid to avoid the disinfectants. It is therefore important that appropriate contact is made between the viroid and the disinfectant if there is to be an effect.

Monitoring and testing

Regular monitoring of a crop allows for the early identification of symptoms. Testing also provides a means of detecting PSTVd in symptomatic, but also asymptomatic, plants, such as ornamentals.

Recovery

Potato spindle tuber viroid-free plants can be recovered from the infected plants by first exposing them to low temperatures (5-8°C) and then producing a meristem culture from these plants. This resulted in a recovery rate of 18.5-80% (Lizarraga *et al.* 1980; Paduch-Cichal and Kryczyński, 1987). Treatment of plants at high temperature (33-36°C) and producing a culture from the axillary buds also resulted in PSTVd-free plants, though this only produced a recovery rate of 2.4-6% (Stace-Smith and Mellor, 1970).

Phytosanitary measures

Import control measures

At ports, general surveillance is carried out, involving visual inspection for PSTVd symptoms, and subsequent testing if symptoms are found. If PSTVd is confirmed, the consignment is generally destroyed by either incineration or deep burial. Specific surveys are also instigated if there is considered to be sufficient risk of PSTVd spread.

Impacts

Economic impact

Reported yield losses for potato vary between 10 and 74% (Singh *et al.*, 1971; Cui *et al.*, 1992; Leontyeva, 1963; Cammack and Richardson, 1963; Folsom and Schultz, 1924; Bonde *et al.*, 1943; Murphy *et al.*, 1966; Hunter and Rich, 1964; Martin, 1924, 1928; Wedgworth, 1928; McKay and Dykstra, 1932; Burger, 1927; Balashev, 1941). The level of loss is dependent on the cultivar of potato, the strain of PSTVd and the length of time the

crop has been infected with the viroid. For example, in the Saco cultivar, tuber yield was reduced by 24% when infected with the mild strain, but by 64% when infected with the severe strain (Singh *et al.*, 1971).

Tomato also suffers from variable yield losses (Verhoeven *et al.*, 2004, 2007). As well as the factors mentioned for potato, the growth stage at which the plant is infected is also important. Because fruit stops developing once stunting sets in, if tomato plants are infected prior to fruit production, yield losses could be as much as 100%, whereas if they are infected post fruit production, the fruit may still develop to a marketable size. Further, high infection rates in the past have been caused by a delay in identifying PSTVd in the crop and instigating measures. It has recently been found that elevated ozone concentration can reduce the impact of PSTVd in tomato (Abraitiene and Girgždiene, 2013).

Soliman (2012) calculated the economic losses of PSTVd in Europe if left unchecked to be 4.4 million euros in potato and 5.7 million euros in tomato, with the bulk of the costs borne by consumers having to pay a higher price for the same product, due to decreases in supply. Owens and Verhoeven (2009) have also calculated yield losses in North America over the period 1922-2009 to be 1% even after attempting to control the viroid.

In peppers, ornamentals, and other plants, no yield losses have been recorded, with only mild symptoms in pepper to date (Lebas *et al.*, 2005).

Environmental impact

No impact has been recorded.

Social impact

No impact has been recorded.

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