

Tomato Chlorotic Dwarf Viroid on Greenhouse Tomatoes

March 2021

Tomato chlorotic dwarf viroid (TCDVd) is a Pospiviroid which has a circular single stranded RNA molecule with 360 nucleotides and lack of a protein coat. The absence of protein coat distinguishes viroids from viruses. Viroids are the smallest "organisms" known to cause plant diseases. TCDVd is closely related to but different from the known Potato spindle tuber viroid (PSTVd), the first viroid to be identified. TCDVd shares only 86-88% sequence homology with PSTVd. In relation to crop plants, very little is known about the epidemiology, host specificity and host-pathogen interaction of TCDVd. However, TCDVd is expected to exhibit pathogenicity properties similar to that of PSTVd. In general, pospiviroids use the plant cell nucleolus to self-replicate (i.e. multiply in number) and accumulate. The movement of viroids within an infected plant is via plasmodesmata, from cell to cell, and via phloem throughout the plant. In general, viroids have been known to infect both herbaceous and woody plants, including many ornamentals, but many of them are symptomless carriers.

TCDVd is known to occur in field and greenhouse-grown tomatoes in Europe. In Canada, it was first detected in greenhouse tomatoes, var. "Trust", in Manitoba in 1996. It was successfully eradicated and has not been reported since then. In 2007, TCDVd was reported on tomato from two greenhouses in Arizona. TCDVd was also reported on petunia and verbena but no visible symptoms were observed (source: J.Th.J. Verhoeven, The Netherlands).

Disease Symptoms

The symptoms on infected tomato plants vary depending on the type of TCDVd strain, tomato variety, age of the plant, plant vigour and climatic conditions. Initial symptoms appear as reduced growth and yellowing (chlorosis) of leaves in the young terminal parts of the plant, 3-6 weeks after the infection (Figure IA). With time, the infected plants may become stunted, chlorosis may become more pronounced or turn to bronzing and/or purpling and leaves may show necrotic lesions and become brittle and distorted. The spread of the viroid can readily be seen within a row. The commonly observed symptoms are stunting, overall bunchiness, reduced leaves and fruit, leaf chlorosis, leaf and petiolar necrosis, downward bending of leaves, fruit distortion and even death of plants (Figure IB), similar to the symptoms expressed by PSTVd when artificially inoculated to tomato plants (source: Dr. Rudra Singh). A tomato plant can be infected with both TCDVd and PSTVd simultaneously and, thus, becomes difficult to distinguish one viroid from the other by the symptoms.

Detection of TCDVd

Detection of viroid infection by symptoms is challenging since many viroids and viruses can produce similar symptoms on tomato plants. Unlike viruses, ELISA (enzyme-linked-immunosorbent assay) technique cannot be used for detecting viroids since they lack a protein coat. TCDVd can only be

detected by a molecular PCR-based technique called reverse transcription PCR (RT-PCR) using pospiviroids-specific molecular probes as described by Bostan et al. 2004 and Singh et al., 2006. Note: B.C. Ministry of Agriculture, Food and Fisheries - Plant Diagnostic Laboratory has the RT-PCR diagnostic tool available for testing suspected tomato samples for Pospiviroids.





Figure 1. Tomato plants infected with *Tomato chlorotic dwarf viroid*. Early symptoms of chlorosis of leaves and stunted terminal growth (A) and advanced symptoms of stunted plants with reduced foliage and severe deformation, curling, and chlorosis of leaves (B).

Infection & Spread

Generally, all pospiviroids, including TCDVd, can easily be transmitted mechanically and spread by contact with contaminated pruning tools, farm equipment, clothing, crop handing, and contact between neighbouring plants. Viroids can also be transmitted by seeds, vegetative propagation, pollen, grafting and insects. Although transmission of TCDVd by seeds is strongly suspected, TCDVd was not detected in seeds obtained from TCDVd-infected tomato plants (source: Dr. Rudra Singh, personal communication). However, in potatoes, PSTVd is known to be transmitted by seeds. Since TCDVd is known to infect other solanaceous plants and herbaceous ornamentals the possibility of seed transmission of TCDVd needs further investigation. Transmission of TCDVd by pollen or insects is not yet known. Most efficient transmission of TCDVd is expected when young growing terminal parts of the plant are being handled during pruning and training. Least efficient transmission is expected during picking of fruits and removing old leaves (source: J.Th.J. Verhoeven). TCDVd is active at higher temperatures and thus high rate of viroid-transmission is expected under warm conditions.

Eradication & Management of Viroids

- Use exclusion and eradication strategies to prevent the introduction and spread of TCDVd.
- Enforce strict biosecurity and phytosanitary measures as appropriate for each greenhouse operation. For details, refer to the Ministry's factsheets on "Disinfection and Sanitation Practices"
- Maintain strict hygiene practices (e.g. hand wash, foot dip, avoid bring green plant materials and food items into a greenhouse facility).
- Use viroid-free seeds and transplants; purchase planting stock from reliable sources that produce transplants from viroid-free certified seeds. Facilities that produce tomato transplants must be located in viroid-free areas.

- Carry out periodic and rigorous scouting for virus and viroid-like symptoms during crop production.
- Submit any suspected plant samples to the <u>Ministry of Agriculture</u>, <u>Food and Fisheries Plant Health Laboratory</u> for viroid/virus confirmation.
- Healthy-looking herbaceous ornamental plants and weeds can be asymptomatic carriers of TCDVd as well as other potential tomato viroids (e.g. PSTVd, Columnea latent viroid, Tomato apical stunt viroid, Tomato planta macho viroid) and viruses. Therefore, practice a systematic weed control program and avoid having ornamental plants in and around greenhouses.
- Avoid using contaminated pruning shears and knives. Sterilize pruning tools before and after
 pruning each plant (details on sterilization techniques is attached). Pruners must be sterilized
 thoroughly at each use. Use a couple of pruners alternatively; this will give sufficient time
 (immerse minimum of 2 min in a disinfectant) to disinfect a pruner while pruning with the other.
- Control or minimize movement of people and contact with or handling of infected plants.
- Remove plant debris and infected plants, including root system and plant growth medium; deep bury or incinerate. Since viroids can persist in the root system of infected plants do not reuse the planting bags or containers from the previous crop. TCDVd has a latent period of 3-6 weeks before any symptoms can be seen, therefore symptomless plants surrounding the infected plant must also be destroyed.
- Carry out a thorough greenhouse clean up between crop cycles and at the year-end.
- Pepino mosaic virus control strategies can be adopted in controlling TCDVd.

Useful websites

Emerging Virus Diseases of Greenhouse Vegetable Crops (B.C. Ministry of Agriculture, Food and Fisheries)

<u>Management of Pepino mosaic virus in greenhouse tomato</u> (Ontario Ministry of Agriculture, Food and Rural Affairs)

For Further Information

Boonham N, Gonzáles Pérez L, Mendez M.S, Lilia Peralta E, Blockly A, Walsh K, Barker I and Mumford R.A (2004). Development of a real-time RT-PCR assay for the detection of *Potato spindle tuber viroid*. Journal of Virological Methods 116, 139-146.

Bostan H, Nie X and Singh, R.P (2004). An RT-PCR primer pair for the detection of Pospivirod and its application in surveying ornamental plants for viroids. Journal of Virological Methods 116: 189-193.

Diener, T (1991). Subviral pathogens of plants: viroids and viroid like satellite RNAs. The FASEB Journal 5: 2808-2813.

Hammond, R.W. and Owens, R.A. (2006). Viroids: New and continuing risks for horticultural and agricultural crops. APSnet Feature, November 2006.

Shamloul AM, Hadidi A, Zhu SF, Singh RP and Sagredo B (1997) Sensitive detection of potato spindle tuber viroid using RT-PCR and identification of a viroid variant naturally infecting pepino plants. Canadian Journal of Plant Pathology 19: 89-96.

Singh R.P and Dilworth A.D. (2006) Detection of *Citrus exocortis viroid*, *Iresine viroid* and *Tomato chlorotic dwarf viroid* in new ornamental host plants in India. Plant Disease 90: 1457.

Singh R.P, Dilworth A.D., Singh M. and Babcock, K.M. (2006). An alkaline solution simplifies nucleic acid preparation for RT-PCR and infectivity assays of viroids from crude sap and spotted membrane. Journal of Virological Methods 132: 204-211.

Singh R.P, Xianzhou N and Singh M (1999) *Tomato chlorotic dwarf viroid*: an evolutionary link in the origin of pospiviroids. Journal of General Virology 80: 2823-2828.

Verhoeven, J.Th.J, Jansen, C.C.C, Werkman, A.W. and Roanhorse, J.W. (2007) First report of *Tomato chlorotic dwarf* viroid in *Petunia hybrida* from the United States of America. Plant Disease 91, 324.

Verhoeven, J.Th.J, Jansen, C.C.C, Willemen, T.M, Kox, L.F.F, Owens, R.A and Roanhorse, J.W. (2004) Natural infections of tomato by *Citrus exocortis viroid, Columnea latent viroid, Potato spindle tuber viroid* and *Tomato chlorotic dwarf viroid.* European Journal of Plant Pathology 110: 823-831.

Prepared by:
Siva Sabaratnam, Plant Pathologist
Abbotsford Agriculture Centre,
British Columbia Ministry of Agriculture, Food and Fisheries
Abbotsford, B.C.