



## November 2019 – update - PEST Report (follow-up pest report of October 2019)

### 1.1 Official confirmation of Tomato brown rugose fruit virus (ToBRFV) in *Solanum lycopersicum* at two professional fruit production companies (closed conditions).

#### 1.2 Executive summary

This report concerns the official confirmation of ToBRFV at two fruit production companies of *Solanum lycopersicum* in the Netherlands. The first finding was officially confirmed on October 17 2019.

Following the first suspicion, national measures for ToBRFV were implemented on October 4<sup>th</sup> 2019, as based on the measures of EU implementing decision 2019/615 which entered into force on 1 November 2019.

Extensive investigation of all possible sources of the infections as well as an intensified survey was initiated following the first finding. This has resulted in one more finding as confirmed on 1 November 2019. Furthermore to date 14 suspicions are recorded as based on realtime PCR testing results at other tomato fruit growers and awaiting confirmation as well as several indications of other possible suspicions.

To date 48 tomato fruit companies have been thoroughly investigated out of 89 targeted companies.

The source of the outbreaks is not known. Further tracing investigations of plant growers and seed companies are on-going.

The organism is regulated as part of EU Commission implementing decision 2019/1615. Identity of the pest (scientific name) Tomato brown rugose fruit virus.

Categorization of the pest EU Commission implementing decision 2019/1615.

Location: municipality Westland (2 findings).

Reason of the notification: Update report.

How the pest was found (1) pest related official survey;

Information on the infested area, severity and source of the outbreak – summary

At the first company visual symptoms were observed on approximately 8% of plants in a fruit production site of 2.8 ha. Fruits of affected plants showed a delay in ripening.

At other companies damage is variable, ranging from limited damage to similar impact as for the first company. Total infested area (11.8 ha – two production sites); Suspicion (real-time PCR positive): 14 production places or sites totaling 344.5 ha.

Official phytosanitary measures - summary

Measures are aimed at preventing new introductions and spread of ToBRFV. At affected fruit production sites, strict hygiene measures are applied including restricting access, disinfection or replacement of clothing, machines, equipment, surfaces and packaging

<p>material. In particular use of the disinfection agent potassium peroxymonosulfate is recommended.</p> <p>For any fruits harvested from the production place, specific hygiene measures are applied both at the fruit production place as well as the packing station, including cleaning and disinfection of packaging material.</p> <p>Following removal of the crop, cleaning and disinfection of the greenhouse, the production site will be monitored, including testing, to verify absence of the virus in the succeeding crop.</p> <p>Specific surveillance has been intensified targeting fruit companies based on information from fruit growers, packaging stations, plant growers and private labs.</p>	
<b>1.3 Type of notification</b>	(2) full notification (second notification within 30 days)
<b>2.1 Single Authority</b>	Notification from the National Plant Protection Organization of the Netherlands – Netherlands Food and Consumer Product Safety Authority
<b>2.2 Official contact</b>	M.B. de Hoop. +31651584878 Email: <a href="mailto:m.b.dehoop@nvwa.nl">m.b.dehoop@nvwa.nl</a>
<b>3. Location of presence of harmful organism</b>	municipality Westland (2 findings)
3.2 Map of the location.	
<b>4. Reason of the notification and pest status</b>	(1) First presence of the harmful organism
<b>4.3 Previous Pest status</b>	(8) Absent: no pest records
<b>4.4 Current Pest status</b>	(15) Transient: actionable, under eradication.
<b>5. Information relating to the finding.</b>	(1) pest related official survey.
5.2 Date of finding.	<p>Suspicious symptoms at the production site were observed on October 1, 2019.</p> <p>The suspicion of ToBRFV was confirmed by DAS-ELISA, bio-assay and real-time PCR on 7 October 2019.</p> <p>Full confirmation by NGS (Next Generation Sequencing) was determined on October 17 2019.</p>
5.3 Sampling for laboratory analysis	Leaf samples of the infected plant were analysed by the accredited laboratories of NPPO and Naktuinbouw and tested positive in DAS-ELISA, bio-assay and real-time PCR (ISHI test protocol).
5.4 Laboratory	<p>Mr Maikel Aveskamp</p> <p>Tel: +31 65 124 7175 Email: <a href="mailto:m.m.aveskamp@nvwa.nl">m.m.aveskamp@nvwa.nl</a></p>

	National Reference Centre - NPPO of the Netherlands
5.5 Diagnostic method.	See 5.3 Leaf samples were screened for ToBRFV by DAS-ELISA and bio-assay at the laboratory of the NPPO. Additionally, leaf material of the same sample, was sent to the Laboratory of Naktuinbouw for confirmation by real-time RT-PCR adopted from International Seed Federation protocol for "Detection of infectious tomato brown rugose fruit virus in Tomato and Pepper seed", September 2019. ( <a href="https://www.worldseed.org/wp-content/uploads/2019/09/Tomato-ToBRFV_2019.09.pdf">https://www.worldseed.org/wp-content/uploads/2019/09/Tomato-ToBRFV_2019.09.pdf</a> ). Definite identification of ToBRFV through next generation sequencing followed by sequencing analysis (of nearly complete genome) on material of the original sample plant.
5.6 Date of official confirmation of the harmful organism's identity	The suspicion of ToBRFV was confirmed by DAS-ELISA and realtime PCR testing results on 7 October 2019. Definite identification of ToBRFV was determined on 17 October 2019.
<b>6. Information related to the area, severity of the finding and source of the finding</b>	(1) Two production sites - infested area (11.8 ha); 8 % infested plants Suspicion (real-time PCR positive): 14 production places or sites totaling 344.5 ha
6.2. Characteristics of the infested area and its vicinity.	(3) Physically closed conditions (3.1) greenhouse; Plants for planting for fruit production of <i>Solanum lycopersicum</i> .
6.3. Host plants in the infested area and its vicinity.	There are many professional fruit production companies of <i>Solanum lycopersicum</i> en <i>Capsicum</i> spp. in the immediate vicinity.
6.4. Infested plant(s), plant product(s) and other object(s).	Fruit production company of <i>Solanum lycopersicum</i> .
6.5. Vectors present in the area.	Bumble bees for pollination in greenhouses.
6.6. Severity of the outbreak.	Visual symptoms were observed on approximately 8% of plants in a fruit production site of 2.8 ha. Some fruits of affected plants showed a delay in ripening. Some plants are also infected with Pepino mosaic virus. Symptoms may also be caused by this virus or a mixed infection of this virus together with ToBRFV. At other companies damage is variable, ranging from limited damage to similar impact as for the first company.



Figure 1: Some fruits of ToBRFV and PepMV-affected tomato plants showed a delay in ripening.  
(NPPO of the Netherlands, 20191007)



Figure 2: Mosaic symptoms in the young leaves of ToBRFV and PepMV-affected tomato plants  
(NPPO of the Netherlands, 20191007)

	 <p>Figure 3: Narrowing of young leaves, typical symptoms of several tobamoviruses in tomato plants. (NPPO of the Netherlands, 20191007)</p>
<p>6.7. Source of the outbreak.</p>	<p>The source of the outbreaks is not known. Further tracing investigations of plant growers and seed companies are on-going.</p>
<p><b>7. Official phytosanitary measures</b></p>	
<p>7.1. Adoption of official phytosanitary measures.</p>	<p>(1) Official phytosanitary measures in the form of chemical, biological or physical treatment, have been taken; At the fruit production site, strict hygiene measures are applied including restricted access and phytosanitary safe removal and destruction of any waste. Machines and other equipment may only leave the location after cleaning and disinfection. Packaging material used for fruits harvested from the production place should be cleaned and disinfected before it is returned to production locations. Packaging stations should follow a strict protocol to avoid any reinfestation of packaging material including cleaning and disinfection of transport vehicles and strict hygiene measures for staff. In particular use of the disinfection agent potassium peroxymonosulfate is recommended. Following removal of the crop, cleaning and disinfection of the greenhouse, the production site will be monitored,</p>

	including testing, to verify absence of the virus in the succeeding crop. Specific surveillance has been intensified targeting fruit companies based on information from fruit growers, packaging stations, plant growers and private labs.
7.2. Date of adoption of the official phytosanitary measures. In case of temporary measures, indication of their expected duration.	4 October 2019.
7.4. Objective of the official phytosanitary measures.	(1) eradication;
7.5. Measures affecting the movement of goods. Indication of one of the following options	(1) measures affect import into or movement within the Union of goods; In the case of option (1), description of the measures. As of 4 October 2019, every suspicion of the presence of the virus should be reported by professional operators (including laboratories) to the NPPO. National measures will be implemented as of this date in line with EU Implementing Decision 2019/1615.
7.6. Specific surveys.	Specific surveillance has been intensified targeting fruit companies based on information from fruit growers, packaging stations, plant growers and private labs. This has resulted in one more finding as confirmed on 1 November 2019. Furthermore to date 14 suspicions are recorded as based on realtime PCR testing results at other tomato fruit growers and awaiting confirmation as well as several indications of other possible suspicions. To date 48 tomato fruit companies have been thoroughly investigated out of 89 targeted companies.
<b>8. Pest risk analysis/assessment</b>	(1) Pest risk analysis is not required (harmful organism is listed in Annex I or Annex II of Directive 2000/29/EC, or is subject to measures adopted pursuant to Article 16(3) of that Directive);

<b>9. Links to relevant websites, other sources of information.</b>	<a href="https://english.nvwa.nl/topics/pest-reporting/contents/pest-reports">https://english.nvwa.nl/topics/pest-reporting/contents/pest-reports</a>
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