



June 2024 PEST Report - THE NETHERLANDS

1.1 First finding of American plum line pattern virus (APLPV) on 11 trees of *Prunus* in two reference (living plant) collections of the NPPO.

1.2 Executive summary

This report concerns the official finding of APLPV in two different reference (living plant) collections for preservation of viruses in *Prunus*. At one location this concerns two individual *Prunus* trees of approximately 10 years old (*Prunus x yedoensis* and *Prunus serrulata*) and at the other location 9 *Prunus* trees (*Prunus subhirtella*, *Prunus x yedoensis*, *Prunus avium* and *Prunus serrulata*). The identity of APLPV was confirmed on 30 May 2024 by the National Reference Laboratory. The two trees at one location had been sampled earlier in 2022 as part of an extensive categorisation project using High Throughput Sequencing (HTS) for testing the entire collection of reference plant material of the NPPO. Fourteen other *Prunus* trees in the same and neighbouring row tested negative for APLPV, based on individual sampling and testing of each tree. The other location was visited since the infected plants at the first location are clonally linked to the plants at the second location.

This reference plant material is maintained as a resource of the virus collection of the NPPO of the Netherlands and used as positive controls, test development, ring testing and knowledge development.

In total three different genome sequence types of the virus were detected, suggesting more than one source. Although the exact source remains uncertain, the three virus genotypes may have been present in some of the accessions or clonal lines for decades (see 6.7). Screening with real-time RT-PCR and sequence analyses indicate limited spread has taken place at both locations. At location 1 the virus seems to have spread from one *Prunus x yedoensis* tree to a neighbouring *P. serrulata* tree. At location 2 it spread from one *Prunus x yedoensis* accession (consisting of 2 trees) to neighbouring trees and from two *Prunus avium* accessions to neighbouring trees. Since the only known transmission pathway for this virus is via propagation material, transmission may have occurred through root contact.

No typical symptoms were observed on the trees. The trees will be destroyed. Neighbouring trees that tested negative, will again be tested in 2025.

The organism is listed as a quarantine pest in EU regulation 2019/2072.

Identity of the pest (scientific name) American plum line pattern virus

Categorization of the pest EU quarantine pest

Location: municipality, city or province.

Reason of the notification: First report / Updated situation / Eradication / New pest ...)


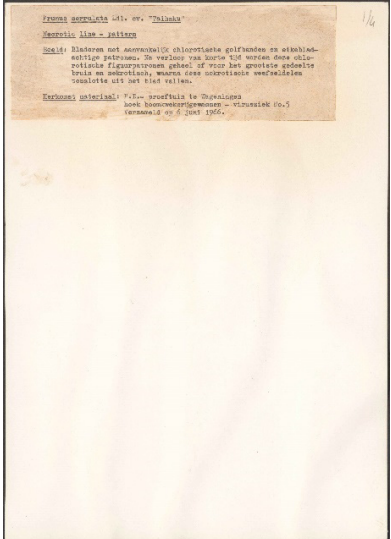
How the pest was found (e.g. (6) information submitted by professional operators, laboratories or other persons;

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

No typical symptoms were observed on the trees.

<u>Official phytosanitary measures</u> - summary	
The trees will be destroyed. Other negatively tested neighbouring trees, will again be tested in 2025.	
1.3 Type of notification	Full notification
2.1 Single Authority	Notification from the National Plant Protection Organization of the Netherlands – Netherlands Food and Consumer Product Safety Authority
2.2 Official contact	M.B. de Hoop. +31651584878 Email: m.b.dehoop@nvwa.nl
3. Location of presence of harmful organism	Municipality: Wageningen and "Horst aan de Maas".
3.2 Map of the location.	Not relevant
4. Reason of the notification and pest status	(1) First presence of the harmful organism
4.3 Previous Pest status	(8) Absent: no pest records
4.4 Current Pest status	(5) Present: localized, under eradication;
5. Information relating to the finding.	5.1 How the harmful organism was found. (6) information submitted by professional operators, laboratories or other persons;
5.2 Date of finding.	30 May 2024.
5.3 Sampling for laboratory analysis	The identity of APLPV was confirmed on 30 May 2024 by the National Reference Laboratory. The two trees at one location have been sampled earlier in 2023 as part of an extensive categorisation project using High Throughput Sequencing (HTS) for testing the entire collection of reference plant material of the NPPO. From each tree a separate sample was taken for testing, consisting of flower or leaf buds from at least three different branches per tree.
5.4 Laboratory	Detection test: Naktuinbouw Confirmation test: National Reference laboratory - NPPO of the Netherlands

	<p>Netherlands Institute for Vectors Invasive plants and Plant health (NIVIP) Contactperson: Jos Kesseleer (j.m.a.kesseleer@nvwa.nl, +31 625560150)</p>
5.5 Diagnostic method.	<p>Detection (Naktuinbouw): Real-time RT-PCR Confirmation and identification (NRL-NIVIP): High Throughput Sequencing (HTS).</p> <p>Samples were first screened with a validated real-time RT-CR for detection of APLPV by Naktuinbouw. Positive samples were subsequently tested with the validated Illumina Sequencing (RNAseq) test, resulting in the complete genome sequences of APLPV.</p>
5.6 Date of official confirmation of the harmful organism's identity	30 May 2024
6. Information related to the area, severity of the finding and source of the finding	<p>Two infested plants for planting of <i>Prunus</i> (pieces). (<i>Prunus x yedoensis</i> and <i>Prunus serrulata</i>).</p> <p>Nine <i>Prunus</i> trees (1 x <i>Prunus subhirtella</i>, 2 x <i>Prunus x yedoensis</i>, 5 x <i>Prunus avium</i> and 1 x <i>Prunus serrulata</i>).</p>
6.2. Characteristics of the infested area and its vicinity.	<p>(2) Open air – other (2.5) other, with specification of the particular case.</p> <p>Reference material for the virus collection of the NPPO of the Netherlands and used as positive controls, test development, ring testing and knowledge development.</p>
6.3. Host plants in the infested area and its vicinity.	<p>Two rows of in total 16 <i>Prunus</i> plants (Wageningen). Several rows of <i>Prunus</i> plant at "Horst aan de Maas".</p>
6.4. Infested plant(s), plant product(s) and other object(s).	<p>Two infested plants for planting of <i>Prunus</i> (pieces). (<i>Prunus x yedoensis</i> and <i>Prunus serrulata</i>).</p> <p>Nine <i>Prunus</i> trees (1 x <i>Prunus subhirtella</i>, 2 x <i>Prunus x yedoensis</i>, 5 x <i>Prunus avium</i> and 1 x <i>Prunus serrulata</i>).</p>
6.5. Vectors present in the area.	Not relevant. The virus is not transmitted by vectors.

6.6. Severity of the outbreak.	No symptoms observed.
6.7. Source of the outbreak.	<p>The origin of the finding is uncertain, but the virus may have been present for decades.</p> <p>The (3) sources may be related to a previous reference (living plant) collection maintained from the 1960s in a.o. Wageningen. Part of the plants/trees from this reference collection were included in the historic herbarium collection in storage at the NPPO (NIVIP Collections).</p> <p>During another extensive sequencing project, dried leaf material from the historic herbarium collection was analyzed using High Throughput Sequencing (HTS). In a <i>Prunus serrulata</i> specimen, which was included in the herbarium in 1966 and at that time was growing in the reference collection in Wageningen, the American plum line pattern virus (APLPV), Cherry virus A, and Little cherry virus 2 were identified (see link). The near-complete APLPV genome sequence obtained shows close similarity to one of the three sequence variants identified in 2024. Further analyses of <i>Prunus</i> samples from the herbarium will be performed to assess if these may have been a source or in any other way related to the current findings.</p> <div style="display: flex; justify-content: space-around; align-items: center;">   </div>
7. Official phytosanitary measures	

7.1. Adoption of official phytosanitary measures.	(3) Official phytosanitary measures will be taken; Destruction. Prior to destruction, valuable plant material will be transferred to quarantine greenhouse facilities for future research and access to collection material.
7.2. Date of adoption of the official phytosanitary measures. In case of temporary measures, indication of their expected duration.	30 May 2024.
7.3. Identification of the area covered by official phytosanitary measures — indicate the method used to identify the area covered by official phytosanitary measures. Provide the results of the surveys that have been carried out.	11 trees
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	(2) measures do not affect import into or movement within the Union of goods.
7.6. Specific surveys.	Other negatively tested neighbouring trees, will again be tested in 2025.
8. Pest risk analysis/assessment	(1) Pest risk analysis is not required (harmful organism is listed in Annex II of Regulation 2019/2072, or is subject to measures adopted pursuant to Article 30 of Regulation 2016/2031).
9. Links to relevant websites, other sources of information.	https://english.nvwa.nl/topics/pest-reporting/contents/pest-reports

