EUROPEAN AND MEDITERRANEAN PLANT PROTECTION ORGANIZATION
ORGANISATION EUROPEENNE ET MEDITERRANEENNE POUR LA PROTECTION DES PLANTES
Summary sheet of validation data for a diagnostic test

The EPPO Standard PM 7/98 *Specific requirements for laboratories preparing accreditation for a plant pest diagnostic activity* describes how validation should be conducted. It also includes definitions of performance criteria.

Laboratory contact details	Walloon Agricultural Research Centre (CRA-W) Département Sciences du Vivant Unité Santé des plantes et forêts Bâtiment Marchal Rue de Liroux, 4, 5030 Gembloux, Belgium
Short description of the test	Detection and identification of Polerovirus by Molecular Conventional RT PCR in Leaves
Date, reference of the validation report	2025-11-13 - Generic detection and identification of Polerovirus
Validation process according to EPPO Standard PM7/98?	yes
Is the lab accredited for this test?	no
Was the validated data generated in the framework of a project?	Other_project
If yes, please specify	VIROBETT project (PRW 204/3, grant -D65-1429)
Description of the test	
Organism(s)	Polerovirus (1POLVG)
Detection / identification	detection and identification
Matrix(ces) tested	Leaves
Plant species tested	Avena sativa, Beta vulgaris, Brassica napus, Capsella bursa-pastoris, Capsicum annuum, Cucumis sativus, Cucurbita pepo, Nicotiana tabacum, Raphanus sativus, Solanum lycopersicum, Solanum tuberosum, Ullucus tuberosus
Method(s)	Molecular Conventional RT PCR
Method: Molecular Conventional RT PCR	
Reference of the test description	
As or adapted from an EPPO diagnostic protocol	no
New test being considered for inclusion in the next version of the EPPO diagnostic protocol?	yes
As or adapted from an IPPC diagnostic protocol	no
Reference of the test	Knierim, D., Tsai, W.S., Kenyon, L., 2013. Analysis of sequences from field samples reveals the

	presence of the recently described Pepper vein yellows virus (genus Polerovirus) in six additional countries. Arch. Virol. 158, 1337–1341.
Is the test modified compared to the reference test	yes Primers Gen1 and Gen2 were used for RT-PCR using Titan One Tube RT-PCR System
Kit	
Is a kit used	no
Other information	
Reaction type	Simplex
Other details on the test	RT-PCR using the generic primers Gen1/2 (Knierim et al., 2013) was performed using the Titan One Tube RT-PCR System (Roche) in 20 µL reaction containing Titan reaction buffer 5X, 500 nM of each primers, dNTPs at 10 nM each, and 0.4 µL of enzyme mix. Thermocycling conditions consisted of a first cycle at 50°C for 30 min for the RT followed by 2 min at 94°C and then 40 cycles of 94°C for 15 s, 54°C for 30 s and 72°C for 45 s. A final elongation step of 5 min at 72°C ended the run.
Performance Criteria :	
Organism 1.:	**Polerovirus(1POLVG)**
Analytical sensitivity	
What is the smallest amount of target that can be detected reliably?	Up to 10^3-fold dilution of the starting material infected with BWYV isolate (P6-6D-W11_BWYV)
Diagnostic sensitivity	
Proportion of infected/infested samples tested positive compared to results from the standard test, see appendix 2 of PM 7/98	100% for the polerovirus (BWYV, BChV, BMYV and TuYV) that were compared with ELISA
Standard test(s)	BWYV TAS-ELISA (RT-0049) kit from DSMZ
Analytical specificity - inclusivity	
Number of strains/populations of target organisms tested	BWYV, BChV, BMYV, TuYV, PLRV, CYDV-RPV, PeVYV, CABYV, UPolV, BLYV
Specificity value	100%
Analytical specificity - exclusivity	
Number of non-target organisms tested	BYV, BtMV
Specificity value	100%
Cross-reacts with	Polerovirus
Diagnostic Specificity	
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	100%
Specify the test(s)	BWYV TAS-ELISA (RT-0049) kit from DSMZ
Reproducibility	

Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100%
Repeatability	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100%
Test performance study	
Test performance study?	no

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