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	Naktuinbouw Sotaweg 22, 2371 GD Roelofarendsveen, Netherlands	
Short description of the test	Real-time RT-PCR (TaqMan RT-PCR) for Potato spindle tuber viroid (PSTVd) and/or Tomato chlorotic dwarf viroid (TCDVd) in leaf material of horticultural crops	
Date, reference of the validation report	2012-08-28 - v1.2	
Validation process according to EPPO Standard PM7/98?	yes	
Is the lab accredited for this test?	yes	
Was the validated data generated in the framework of a project?		
Description of the test		
Organism(s)	Potato spindle tuber viroid / Pospiviroid fusituberis (PSTVD0) Tomato chlorotic dwarf viroid / Pospiviroid chloronani (TCDVD0)	
Detection / identification	detection	
Method(s)	Molecular Extraction DNA RNA Molecular Extraction DNA RNA (2) Molecular real time RT PCR	
Method: Molecular Extraction DNA RNA		
Reference of the test description		
Kit		
Is a kit used	yes	
Manufacturer name	QIAGEN	
Specify the kit used	RNeasy Plant Mini Kit	
Kit used following the manufacturer's instructions?		
Other information		
Method: Molecular Extraction DNA RNA (2)		
Reference of the test description		
Kit		

Is a kit used	yes	
Manufacturer name	LGC	
Specify the kit used	sbeadex maxi plant	
Kit used following the manufacturer's instructions?		
Other information		
Method: Molecular real time RT PCR		
Reference of the test description		
As or adapted from an EPPO diagnostic protocol	no	
As or adapted from an IPPC diagnostic protocol	yes	
IPPC diagnostic Protocol name	ISPM 27 Annex 07 DP 07: Potato spindle tuber viroid (version 2016)	
Name of the test	Real-time RT-PCR using the primers of Boonham et al. (2004)	
Other information		
Are the performance characteristics included in the EPPO diagnostic protocol?	no	
Performance Criteria :		
Organism 1.:	Pospiviroid fusituberis(PSTVD0)	
Analytical sensitivity		
Analytical sensitivity What is smallest amount of target that can be detected reliably?	Solanum lycopersicon: up to 10^6 - 10^7 dilution in sap of healthy tomato leaves. Ornamentals: Relative sensitivity dependent on initial viroid concentration and host plant species. Validated for bulking rates up to 25 for Brugmansia, Calibrachoa, Dahlia (greenhouse), Petunia, Solanum jasminoides and Streptosolen jamesonii, but test is more sensitive. For some crops like field Dahlia, only the summer period seems suitable for (reliable) testing	
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What is smallest amount of target that can be detected reliably? Analytical specificity - inclusivity Number of strains/populations of target organisms tested Specificity value Analytical specificity - exclusivity Number of non-target organisms tested	in sap of healthy tomato leaves. Ornamentals: Relative sensitivity dependent on initial viroid concentration and host plant species. Validated for bulking rates up to 25 for Brugmansia, Calibrachoa, Dahlia (greenhouse), Petunia, Solanum jasminoides and Streptosolen jamesonii, but test is more sensitive. For some crops like field Dahlia, only the summer period seems suitable for (reliable) testing 9 PSTVd isolates, 5 TCDVd isolates 100% 8 other pospiviroids: CLVd, CSVd, CEVd, IrVd-1, MPVd, PCFVd, TASVd, TPMVd 4 Pospiviroidae: ASSVd, HLVd, HSVd, DLVd 2 avsunviroids: ASBVd, CChMVd 8 viruses (tomato): AMV, CMV, PepMV, PVY, ToMV, TMV, ToCV, TYLCV MPVd, PSTVd and TCDVd are detected equally well (up to 10^6-10^7 dilution in sap of healthy tomato leaves); TMPVd is also detected, but not as well as	

Analytical sensitivity What is smallest amount of target that can be detected reliably? Analytical specificity - inclusivity Number of strains/populations of target organisms tested Specificity value Analytical specificity - exclusivity Number of non-target organisms tested Specificity value Analytical specificity - exclusivity Number of non-target organisms tested Specificity value Analytical specificity - exclusivity Number of non-target organisms tested Specificity value Analytical specificity - exclusivity Number of non-target organisms tested Specificity value MPVd, P (up to 10 leaves); PSTVd, T Reproducibility Provide the calculated % of agreement for a given level of the pest (see PM 7/98)			
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(up to 10 leaves); PSTVd, 7 Reproducibility Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	ospiviroids: CLVd, CSVd, CEVd, IrVd-1, CFVd, TASVd, TPMVd 4 Pospiviroidae: ILVd, HSVd, DLVd 2 avsunviroids: ASBVd, 8 viruses (tomato): AMV, CMV, PepMV, IV, TMV, ToCV, TYLCV		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	TVd and TCDVd are detected equally well 6-10^7 dilution in sap of healthy tomato TMPVd is also detected, but not as well as CDVd or MPVd (up to 10-10^2 dilution).		
given level of the pest (see PM 7/98)	Reproducibility		
Repeatability			
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)			
Test performance study			
Test performance study? no			
Provide the calculated % of agreement for a 100%			

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