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	Institute for Sustainable Plant Protection via Amendola, 122/D, 70126 Bari, Italy	
Short description of the test	detection of Xylella fastidiosa in composite samples of herbaceous hosts	
Date, reference of the validation report	2019-03-13 - POnTE - XF-ACTORS, 2nd Joint Annual Meeting: European Research on Emerging Plant Diseases. Valencia, 23-26 october 2018. Book of abstract: p. 63.	
Validation process according to EPPO Standard PM7/98?	no	
Is the lab accredited for this test?	no	
Was the validated data generated in the framework of a project?	no	
Description of the test		
Organism(s)	Xylella fastidiosa(XYLEFA)	
Detection / identification	detection	
Method(s)	Extraction Molecular Extraction DNA RNA Molecular Extraction DNA RNA (2) Molecular Extraction DNA RNA (3) Molecular real time PCR	
Method: Extraction		
Reference of the test description		
As or adapted from an EPPO diagnostic protocol	yes	
EPPO Diagnostic Protocol name	PM 7/024 Xylella fastidiosa (version 4)	
Is the test modified compared to the reference test	no	
Other information		
Other details on the test	Total DNA were extracted from composite samples of herbaceous plants, prepared as reported in the attached additional file	
Method: Molecular Extraction DNA RNA		
Reference of the test description		

As or adapted from an EPPO diagnostic protocol	yes	
EPPO Diagnostic Protocol name	PM 7/024 Xylella fastidiosa (version 4)	
Is the test modified compared to the reference test	no	
Kit		
Is a kit used	no	
Other information		
Other details on the test	Total DNA were extracted from composite samples of herbaceous plants, prepared as reported in the attached additional file, by using: - CTAB-based protocol;	
Method: Molecular Extraction DNA RNA (2)		
Reference of the test description		
As or adapted from an EPPO diagnostic protocol	no	
As or adapted from an IPPC diagnostic protocol	no	
Kit		
Is a kit used	yes	
Manufacturer name	PROMEGA	
Specify the kit used	Maxwell® RSC PureFood GMO and Authentication Kit	
Kit used following the manufacturer's instructions?	yes proceed as indicated in the manufacture's instruction	
Other information		
Other details on the test	Total DNA were extracted from composite samples of herbaceous plants, prepared as reported in the attached additional file, by using: - "Maxwell® RSC PureFood GMO and Authentication Kit" protocol (Promega)	
Method: Molecular Extraction DNA RNA (3)		
Reference of the test description		
As or adapted from an EPPO diagnostic protocol	yes	
EPPO Diagnostic Protocol name	PM 7/024 Xylella fastidiosa (version 4)	
As or adapted from an IPPC diagnostic protocol	no	
Is the test modified compared to the reference test	no	
Kit		
Is a kit used	yes	
Manufacturer name	QIAGEN	

Kit used following the manufacturer's instructions? Other information Other details on the test Total DNA were extracted from composite samples of herbaceous plants, prepared as reported in the attached additional file, by using:"Modified DNeasy MericonTM Food Standard Protocol" (Qiagen); Method: Molecular real time PCR Reference of the test description As or adapted from an EPPO diagnostic protocol yes EPPO Diagnostic Protocol name PM 7/024 Xylella fastidiosa (version 3) Name of the test Real-time PCR - simplex (Harper et al., 2010; erratum 2013) Is the test modified compared to the reference test no K/t no Simplex - Probe Simplex - Probe Other details on the test gPCR following the condition reported in Appendix S-Realtime PCR (Harper et al., 2010; erratum 2013) in PM 7/24 (3) Are the performance characteristics included in the EPPO diagnostic protocol? no Organism 1: Xylella fastidiosa(XYLEFA) Analytical sensitivity one stem portion of 1,5-2 cm excised from 1 infected yellabl? What is smallest amount of target that can be detected reliably? one stem portion of 1,5-2 cm excised from 1 infected yellabl? Diagnostic sensitivity Proportion of infected/infested samples tested positive compared to results from the standard test, see appendix 2 of PM 7/98 100% f			
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7/24 (3)	Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	100% for each test used to extract the total DNA	
Repeatability	Specify the test(s)		

	1	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100% for each test used to extract the total DNA	
Test performance study		
Test performance study?	no	
Other information		
Any other information considered useful	These validation data were obtained by IPSP-CNR in collaboration with the Department of Soil, Plant and Food Science of the University of Bari (ITAY). For any additional detail, see the attached file. The test was include in the last revised version of PM 7/24 (4), which is in consultation to the NPPO member countries Reference: 2019-03-13 - 2019-03-11 - 2019-03-13 - G. Loconsole, L. Manco, O. Potere, L. Susca, G. Altamura, S. Zicca, D. Boscia, V. N. Savino, M. Saponari, 2018. Implementation of sampling procedures for testing composite samples for Xylella fastidiosa. POnTE - XF-ACTORS, 2nd Joint Annual Meeting: European Research on Emerging Plant Diseases. Valencia, 23–26 october 2018. Book of abstract: p. 63.	
The following complementary files are available online:	 <u>composite samples of herbaceous plants</u> (<u>cabbage</u>) 	

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