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	National Instituite of Biology, Department of Biotechnology and Systems Biology Vecna pot 121, 1000 Ljubljana, Slovenia
Short description of the test	Detection of Xylella fastidiosa by real-time PCR (Francis et al., 2006) in plant material
Date, reference of the validation report	2018-06-14 - Dreo, Tanja, 2018. qPCR for detection of Xylella fastidiosa based on Francis et al., 2006, EJPP 115, 203–213: Review of existing validation data, modification of test and in silico analysis (No. D0013/18). National Institute of Biology, Department of Biotechnology and Systems Biology, Ljubljana; Dreo, Tanja and Pirc, Manca, 2018. qPCR for detection of Xylella fastidiosa based on Francis et al., 2006, EJPP 115, 203–213: Diagnostic specificity and sensitivity determined in spiked samples (PKIe) (No. D0014/18). National Institute of Biology, Department of Biotechnology and Systems Biology, Ljubljana; Dreo, Tanja and Pirc, Manca, 2018. qPCR for detection of Xylella fastidiosa based on Francis et al., 2006, EJPP 115, 203–213: Analytical sensitivity – standard curves (No. D0015/18). National Institute of Biology, Department of Biotechnology and Systems Biology, Ljubljana; Dreo, Tanja and Pirc, Manca, 2018 qPCR for detection of Xylella fastidiosa based on Schaad et al. (2002), Francis et al. (2006), Harper et al., 2010, erratum 2013: Analytical specificity (No. D0027/18). National Institute of Biology, Department of Biotechnology and Systems Biology, Ljubljana.
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?	no
Description of the test	
Organism(s)	Xylella fastidiosa (XYLEFA)
Detection / identification	detection
Method(s)	Molecular Extraction DNA RNA Molecular real time PCR

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 3)
Kit	
Is a kit used	yes
Manufacturer name	BIONOBILE
Specify the kit used	QuickPick™ SML Plant DNA
Kit used following the manufacturer's instructions?	
Other information	
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	Taqman real-time PCR tests (based on Francis et al., 2006)
Is the test modified compared to the reference test	yes See attached document
Other information	
Are the performance characteristics included in the EPPO diagnostic protocol?	**no**
Performance Criteria :	
Organism 1.:	**Xylella fastidiosa(XYLEFA)**
Analytical sensitivity	
What is smallest amount of target that can be detected reliably?	DNA: In total 1000 target copies per mL extracted DNA (log 3 cps/mL as determined with digital PCR) were reliably detected in several X. fastidiosa strains, NIB Z 1962 (X. fastidiosa subsp. multiplex, LMG 9063), NIB Z 1963 (X. fastidiosa subsp. fastidiosa from almond, LMG 15099) and CoDiRo strain. Standard curves in plant material: Concentrations from 5x10^4 to down to 5x10^3 to (target cps/mL) can be reliably detected in samples of olives (10^4), oleander (5x10^3), rosemary (10^4) and lavender (5x10^4) plants tested for latent infection. Spiked PKIe controls: 98 % analytical sensitivity for symptomatic samples (111 different samples of 27 different genera were tested) and 100% analytical sensitivity for asymptomatic samples (66 different samples of 20 different genera were tested).
Diagnostic sensitivity	
Proportion of infected/infested samples	No data available.

tested positive compared to results from the standard test, see appendix 2 of PM 7/98	
Analytical specificity - inclusivity	
Number of strains/populations of target organisms tested	3
Specificity value	100%
Analytical specificity - exclusivity	
Number of non-target organisms tested	90
Specificity value	No cross reactivity.
Diagnostic Specificity	
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	No data available.
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
The following complementary files are available online:	• D0013_18_qPCR_Xyf_Francis_2006_ModificationInSilico • D0014_18_qPCR_Xyf_Francis_2006_DiagnosticSensitivityPKIe • D0015_18_qPCR_Xyf_Francis_2006_AnalyticalSensitivity_SCs • D0027_qPCR_Xyf_HarperSchaadFrancis_AnalyticalSpecificity

Creation date: 2018-10-05 00:00:00 - Last update: 2021-01-22 10:01:05