EUROPEAN AND MEDITERRANEAN PLANT PROTECTION ORGANIZATION
ORGANISATION EUROPEENNE ET MEDITERRANEENNE POUR LA PROTECTION DES PLANTES
(11-17239)

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.

Target Organism	Xylella fastidiosa	
Short description	Detection of Xylella fastidiosa by real-time PCR in plant material	
Laboratory contact details	National Institiute of Biology, Department of Biotechnology and Systems Biology Vecna pot 121, 1000 Ljubljana, Slovenia	
Date and reference of the validation report	2016-02_26 - Dreo, Tanja, 2016. Validation data on the modified real-time PCR for detection of Xylella fastidiosa adapted from Francis et al. (2006) (No. D0002/16). National Institute of Biology, Department of Biotechnology and Systems Biology, Ljubljana.	
Validation process according to EPPO Standard PM 7/98:	No	
Reference of the test description	N/R Supporting data for the new draft of the EPPO diagnostic protocol on Xylella fastidiosa.	
Is the test the same as described in the EPPO DP?		
Is the lab accredited for this test?	No	
Plant species tested (if relevant)	Asparagus acutifolius, Coffea, Lavandula, Nerium oleander, Olea europea, Polygala myrtifolia, Quercus cerris, Rosamrinum officinalis, Spartium junceum.	
Matrices tested (if relevant)	Plant material, mainly mixtures of midribs, petioles and vascular tissues.	
List of methods used		
Method for extraction / isolation / baiting of target organism from matrix		
Molecular methods, e.g. hybridization, PCR and real time PCR	X	DNA extraction from plant material using QuickPick™ SML Plant DNA kit (Bionobile). Modified real-time PCR adapted from Francis, M., Lin, H., Rosa, J.C.-L., Doddapaneni, H., Civerolo, E.L., 2006. Genome-based PCR Primers for Specific and Sensitive Detection and Quantification of Xylella fastidiosa. European Journal of Plant Pathology 115, 203–213. doi:10.1007/s10658-006-9009-4
Serological methods: IF, ELISA, Direct Tissue Blot Immuno Assay		

Plating methods: selective isolation		
Bioassay methods: selective enrichment in host plants, baiting, plant test and grafting.		
Pathogenicity test		
Fingerprint methods: protein profiling, fatty acid profiling & DNA profiling		
Morphological and morphometrical methods intended for identification		
Biochemical methods: e.g. enzyme electrophoresis, protein profiling		
Other		
Analytical sensitivity (= limit of detection)		
What is smallest amount of target that can be detected reliably?	On the DNA extracted from pure cultures of X. fastidiosa: 2.6, 3.2 and 3.5 (log (cells/mL) of Xylella fastidiosa subsp. multiplex, Xylella fastidiosa, and Xylella fastidiosa subsp. pauca CoDiRO strain, respectively. On plant material: 94 % (determined on log 5 cells/mL of plant extracts; the lowest concentration tested)	
Diagnostic sensitivity		
Proportion of infected/infested samples tested positive compared to results from the standard test , see appendix 2 of PM 7/98	No data available.	
Specify the standard test		
Analytical specificity		
Specificity value	100 %	
Number of strains/populations of target organisms tested	4	
Number of non-target organisms tested	15	
Cross reacts with (specify the species)	No cross reactions were observed.	
Diagnostic Specificity		
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	No data available.	
Specify the standard test		
Reproducibility		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	97 %	
Repeatability		

Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	No data avilable.
Test performance study	
Test performance study?	No
Include brief details of the test performance study and its output.It available, provide a link to published article/report	
Other information	
Any other information considered useful **e.g. robustness, ease of performing the test, etc.**	
The following complementary files are available online:	• Validation data on the modified real-time PCR for detection of Xylella fastidiosa adapted from Francis et al. (2006) (No. D0002/16)