

**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.

<b>Target Organism</b>	Xylella fastidiosa	
<b>Short description</b>	Detection of Xylella fastidiosa by real-time PCR in plant material	
<b>Laboratory contact details</b>	National Institute of Biology, Department of Biotechnology and Systems Biology Vecna pot 111, 1000 Ljubljana, Slovenia	
<b>Date and reference of the validation report</b>	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.	
<b>Validation process according to EPPO Standard PM 7/98:</b>	No	
<b>Reference of the test description</b>	N/R Supporting data for the new draft of the EPPO diagnostic protocol on Xylella fastidiosa.	
<b>Is the test the same as described in the EPPO DP?</b>		
<b>Is the lab accredited for this test?</b>	No	
<b>Plant species tested (if relevant)</b>	Asparagus acutifolius, Coffea, Lavandula, Nerium oleander, Olea europea, Polygala myrtifolia, Quercus cerris, Rosamrinum officinalis, Spartium junceum.	
<b>Matrices tested (if relevant)</b>	Plant material, mainly mixtures of midribs, petioles and vascular tissues.	
<b>List of methods used</b>		
<b>Method for extraction / isolation / baiting of target organism from matrix</b>		
<b>Molecular methods, e.g. hybridization, PCR and real time PCR</b>	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
<b>Serological methods: IF, ELISA, Direct Tissue Blot Immuno Assay</b>		

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

<b>Provide the calculated % of agreement for a given level of the pest (see PM 7/98)</b>	No data available.
<b>Test performance study</b>	
<b>Test performance study?</b>	No
<b>Include brief details of the test performance study and its output. If available, provide a link to published article/report</b>	
<b>Other information</b>	
<b>Any other information considered useful e.g. robustness, ease of performing the test, etc.</b>	
The following complementary files are available online:	<ul style="list-style-type: none"> <li>• <a href="#">Validation data on the modified real-time PCR for detection of Xylella fastidiosa adapted from Francis et al. (2006) (No. D0002/16)</a></li> </ul>