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 immunofluorescence in plant material	
Laboratory contact details	Anses, Laboratoire de la Santé des Végétaux - Unité de bactériologie, virologie OGM 7 rue Jean Dixméras, 49044 Angers, France	
Date and reference of the validation report	2015-09-30 - Rapport de caractérisation et de validation de méthode d'analyse - Détection de Xylella fastidiosa par PCR en temps réel sur plantes hôtes MA039	
Validation process according to EPPO Standard PM 7/98:	Yes	
Reference of the test description	N/R In-house antiserum produced in collaboration with INRA Unité de recherche Biopolymères, Interactions, Assemblages – Nantes in 2012	
Is the test the same as described in the EPPO DP?		
Is the lab accredited for this test?	No	
Plant species tested (if relevant)	Vitis vinifera, Prunus persica, Citrus sinensis, Coffea arabica, C. canephora	
Matrices tested (if relevant)	Petioles	
List of methods used		
Method for extraction / isolation / baiting of target organism from matrix		
Molecular methods, e.g. hybridization, PCR and real time PCR		
Serological methods: IF, ELISA, Direct Tissue Blot Immuno Assay	X	Immunofluorescence
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?	- Grapevine (Vitis vinifera): ≈ 10^6 bact./mL - Peach tree (Prunus persica): ≈ 10^5 bact./mL - Coffee tree (Coffea arabica): ≈ 10^4 bact./mL - Coffee tree (C. canephora): ≈ 10^4 bact./mL	
Diagnostic sensitivity		
Proportion of infected/infested samples tested positive compared to results from the standard test , see appendix 2 of PM 7/98	- Grapevine (Vitis vinifera): 40% - Peach tree (Prunus persica): 50% - Orange tree (Citrus sinensis): 78% - Coffee tree (Coffea arabica): 60% - Coffee tree (C. canephora): 30%	
Specify the standard test	- Spiked matrices with bacterial concentration from 10^2 to 10^6 bact./mL - Grapevine spiked with X. f. subsp. fastidiosa (CFBP7970) - Orange tree spiked with X. f. subsp. pauca (CFBP8072) - Peach tree spiked with X. f. subsp. multiplex (CFBP8173 and CFBP8070) - Coffee tree (Coffea arabica) spiked with X. f. subsp. pauca (CFBP8072) - Coffee tree (C. canephora) spiked with X. f. subsp. fastidiosa (CFBP 8074) 20 samples per matrix (orange) 12 samples per matrix (grapevine) 12 samples per matrix (peach tree) 20 samples per matrix (C. arabica) 20 samples per matrix (C. canephora)	
Analytical specificity		
Specificity value	100%	
Number of strains/populations of target organisms tested	Inclusivity tested with 10 target strains: 100% - X.f. subsp. fastidiosa (CFBP8069 -LSV0056/ CFBP8071 -LSV4041/ CFBP8083 -LSV4042/ CFBP7970-LSV2434/ CFBP8082 -LSV4040) - X.f. subsp. pauca (CFBP8072 - LSV4103) - X.f. subsp. sandyi (CFBP8077 -LSV4236) - X.f. subsp. multiplex (CFBP8173 -LSV4039/ CFBP8068-LSV0054/ CFBP8070-LSV4038)	
Number of non-target organisms tested	Exclusivity tested with 17 non-target strains: 100% - 1 Xylophilus ampelinus (CFBP2098) - 2 Xanthomonas arboricola pv. pruni (LSV2574/LSV 2573)	

	- 1 Xanthomonas arboricola pv. juglandis (LSV0862) - 1 Xanthomonas axonopodis pv. citri (LSV2647) - 1 Xanthomonas axonopodis pv. aurantifolia (LSV2680) - 2 Xanthomonas axonopodis pv. phaseoli (LSV1014/LSV3161) - 1 Xanthomonas axonopodis pv. fragariae (LSV3151) - 1 Xanthomonas fragariae (LSV2553) - 1 Xanthomonas hortorum pv. carotae (LSV1776) - 1 Xanthomonas campestris pv. campestris (LSV0455) - 1 Xanthomonas campestris pv. juglandis (LSV1158) - 1 Xanthomonas hortorum pv. hedera (LSV2303) - 1 Xanthomonas translucens pv. graminis (LSV0628) - 1 Xanthomonas translucens pv. hordei (LSV0629) - 1 Xanthomonas oryzae pv. oryzae (LSV0865)
Cross reacts with (specify the species)	None
<u>Diagnostic Specificity</u>	
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	- Coffee tree (Coffea arabica): 100% - Coffee tree (C. canephora): 100%
Specify the standard test	
<u>Reproducibility</u>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	
<u>Repeatability</u>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	- Grapevine (Vitis vinifera): 67% - Peach tree (Prunus persica): 83%
<u>Test performance study</u>	
Test performance study?	No
Include brief details of the test performance study and its output.It available, provide a link to published article/report	
<u>Other information</u>	
Any other information considered useful e.g. robustness, ease of performing the test, etc.	