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	Xanthomonas axonopodis pv. dieffenbachiae	
Short description	Detection of Xanthomonas axonopodis pv. dieffenbachiae by nested-PCR in leaves and pure culture	
Laboratory contact details	Anses Plant Health Laboratory - Pests and Tropical Pathogens Unit Pôle de Protection des Plantes, 7 Chemin de l'IRAT, 97410 Saint Pierre, France	
Date and reference of the validation report	2012-03 - Inter-laboratory ring test : Xanthomonas axonopodis pv. dieffenbachiae in Anthurium (Report Xad01-version 2)	
Validation process according to EPPO Standard PM 7/98:	No	
Reference of the test description	N/R	
Is the test the same as described in the EPPO DP?	Yes	
Is the lab accredited for this test?	No	
Plant species tested (if relevant)	Anthurium sp.	
Matrices tested (if relevant)	Leaves and pure culture	
List of methods used		
Method for extraction / isolation / baiting of target organism from matrix	X	Extraction as in Appendix 1 of PM7/23(2)
Molecular methods, e.g. hybridization, PCR and real time PCR	X	Nested-PCR as in Appendix 4 of PM7/23(2)
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?	1x10^4 CFU.mL-1	
Diagnostic sensitivity		
Proportion of infected/infested samples tested positive compared to results from the standard test , see appendix 2 of PM 7/98	Comparative study : 100% ; Collaborative study : 97.5%	
Specify the standard test	Isolation + AGDIA Indirect-ELISA on pure culture (OEPP PM7/23)	
Analytical specificity		
Specificity value	100%	
Number of strains/populations of target organisms tested	50 (see attached downloadable file Appendix 1)	
Number of non-target organisms tested	53 (see attached downloadable file Appendix 2)	
Cross reacts with (specify the species)	The restriction step performed after the N-PCR enables to exclude all the tested strains. Without the restriction step, a Xanthomonas axonopodis pv. allii strain and some Xanthomonas campestris pv. syngonii strains can not be excluded.	
Diagnostic Specificity		
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	Comparative study : 96% ; Collaborative study : 95%	
Specify the standard test	Isolation + AGDIA Indirect-ELISA on pure culture (OEPP PM7/23)	
Reproducibility		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	93%	
Repeatability		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	94%	
Test performance study		
Test performance study?	Yes	
Include brief details of the test	Results obtained with the N-PCR are excellent for all criteria	

performance study and its output.It available, provide a link to published article/report	(>= 90%) and not statistically different from results obtained with the standard test. The most important difference between the N-PCR and the standard test concerns the analytical sensitivity. Therefore, we proposed maintaining a step of pathogen isolation in the revised EPPO detection scheme.
Other information	
Any other information considered useful **e.g. robustness, ease of performing the test, etc.**	When other criteria besides technical performance are considered, the N-PCR has advantages compared to the other methods tested : -It produces results more quickly (2-3 days) than the reference method for approximately the same cost. -It is easily transferable in comparison to isolation and the IF test, which require experience for recognising the typical bacteria.
The following complementary files are available online:	• Appendix_1-List_target_strains • Appendix_2-List_non_target_strains • Inter-laboratory ring test : Xanthomonas axonopodis pv. dieffenbachiae in Anthurium (Report Xad01-version 2)