## 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	Council for Agricultural Research and Economics- Research Centre for Plant Protection and Certification Via Carlo Giuseppe Bertero, 22, 00156 Rome, Italy
Short description of the test	Intralaboratory validation of Ceratocystis platani_SYBR Green Real-Time PCR in wood of Platanus acerifolia
Date, reference of the validation report	2025-05-04 - Intralaboratory validation of Ceratocystis platani_Real-Time PCR_SYBR Green (corresponding M. Pilotti)
Validation process according to EPPO Standard PM7/98?	yes
Is the lab accredited for this test?	no
Was the validated data generated in the framework of a project?	no
Description of the test	
Description of the test	
Organism(s)	Ceratocystis platani (CERAFP)
Detection / identification	detection and identification
Method(s)	Molecular real time PCR
Method: Molecular real time PCR	
Reference of the test description	
As or adapted from an EPPO diagnostic protocol	no
New test being considered for inclusion in the next version of the EPPO diagnostic protocol?	yes
As or adapted from an IPPC diagnostic protocol	no
Reference of the test	Lumia et al., 2018
Is the test modified compared to the reference test	no
Kit	
Is a kit used	no
Other information	

Reaction type	Simplex	
Other details on the test	SsoAdvanced Universal SYBR Green Supermix (BIO-RAD)	
Performance Criteria :		
Organism 1.:	Ceratocystis platani(CERAFP)	
Analytical sensitivity		
What is smallest amount of target that can be detected reliably?	3 fg of C. platani gDNA per PCR reaction (the detection limit was also confirmed by spiking with 6 ul of an extract from C. platani-uninfected and Fomitiporia mediterranea-infected necrotic wood)	
Diagnostic sensitivity		
Proportion of infected/infested samples tested positive compared to results from the standard test, see appendix 2 of PM 7/98	100%	
Standard test(s)	Comparison with samples of known status	
Reproducibility		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100% - evaluated by two different operators; each operator performed three different experiment testing 8 replicates at the LOD (3 fg)	
Repeatability		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100% - evaluated on 8 replicates at the LOD (3 fg). The result was also confirmed by testing the gDNA of five additional C. platani strains, each in a single experiment performed by one operator, at the limit of detection and with eight replications.	
Test performance study		
Test performance study?	no	
The following complementary files are available online:	• <u>Lumia et al (2018)</u>	

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