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	Naktuinbouw	
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Short description of the test	Detection of Acidovorax citrulli by PCR in seeds	
Date, reference of the validation report	2015-11-16 - 1v1.2	
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		
Organism(s)	Acidovorax citrulli (PSDMAC)	
Detection / identification	detection	
Method(s)	Molecular Extraction DNA RNA Molecular real time PCR	
Method: Molecular Extraction DNA RNA		
Reference of the test description		
As or adapted from an EPPO diagnostic protocol	yes	
EPPO Diagnostic Protocol name	PM 7/127 <i>Acidovorax citrulli</i> (version 1)	
Is the test modified compared to the reference test	no	
Kit		
Is a kit used	yes	
Manufacturer name	LGC	
Specify the kit used	sbeadex maxi plant	
Kit used following the manufacturer's instructions?		
Other information		
Other details on the test	DNA extraction using Kingfisher and Sbeadex maxi kit for Acidovorax citrulli (LGC Genomics)	
Method: Molecular real time PCR		

Reference of the test description		
As or adapted from an EPPO diagnostic protocol	yes	
EPPO Diagnostic Protocol name	PM 7/127 <i>Acidovorax citrulli</i> (version 1)	
Name of the test	Real-time PCR targeting the IS1002 element (Woudt et al., 2009a,b) and Contig 22	
Is the test modified compared to the reference test	no	
Other information		
Are the performance characteristics included in the EPPO diagnostic protocol?	yes	
Performance Criteria :		
Organism 1.:	Acidovorax citrulli(PSDMAC)	
Analytical sensitivity		
What is smallest amount of target that can be detected reliably?	The limit of detection at 0.95 probability is 9 cells/mL seed extract.	
Diagnostic sensitivity		
Proportion of infected/infested samples tested positive compared to results from the standard test, see appendix 2 of PM 7/98	Not determined	
Standard test(s)	No standard test available	
Analytical specificity - inclusivity		
Number of strains/populations of target organisms tested	168 strains	
Specificity value	100%	
Analytical specificity - exclusivity		
Number of non-target organisms tested	54 non-targets	
Specificity value	Two primers sets tested - IS1002 cross-reacts with 2 of the 9 Acidovorax cattleyae isolates tested and several unknown bacteria - Contig22 cross-reacts only with 1 unknown bacteria, characterized in AFLP-study, outside of the Acit tree. Cross-reacts with both primer sets	
Cross reacts with	Acidovorax cattleyae	
Diagnostic Specificity		
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	Diagnostic specificity: 98%	
Specify the test(s)	AFLP-study	
Reproducibility		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100% Intralaboratory testing	
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Repeatability		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100% Inter- and intralaboratory testing	
Test performance study		
Test performance study?	no	
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The following complementary files are available online:	 Poster Koenraadt et al 2014 ISTA Seed Health Symposium Specificity of Contig21 Taqman Validation report Acidovorax citrulli v1.2 	

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