

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	detection of <i>Xanthomonas hortorum</i> pv. <i>gardneri</i> , <i>Xanthomonas euvesicatoria</i> pv. <i>perforans</i> , <i>Xanthomonas vesicatoria</i> , <i>Xanthomonas</i> <i>euvesicatoria</i> pv. <i>euvesicatoria</i> <i>Xanthomonas</i> <i>euvesicatoria</i> pv. <i>euvesicatoria</i> by Molecular real time RT PCR in Seeds
Date, reference of the validation report	2022-02-10 - Validation_ASPROPI_2017_pp143-166
Link to other validation data	- Validation_ASPROPI_2017_pp143-166 detection of <i>Xanthomonas euvesicatoria</i> pv. <i>euvesicatoria</i> , <i>Xanthomonas hortorum</i> pv. <i>gardneri</i> , <i>Xanthomonas</i> <i>euvesicatoria</i> pv. <i>perforans</i> , <i>Xanthomonas</i> <i>vesicatoria</i> <i>Xanthomonas vesicatoria</i> by Molecular Conventional PCR in Seeds
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?	Other_project
If yes, please specify	ASPROPI (Founded by MIPAAF_Italy)
Description of the test	
Organism(s)	<i>Xanthomonas hortorum</i> pv. <i>gardneri</i> (XANTGA) <i>Xanthomonas euvesicatoria</i> pv. <i>perforans</i> (XANTPF) <i>Xanthomonas vesicatoria</i> (XANTVE) <i>Xanthomonas euvesicatoria</i> pv. <i>euvesicatoria</i> (XANTEU)
Detection / identification	detection
Method(s)	Molecular real time RT PCR
Method: Molecular real time RT PCR	
Reference of the test description	
New test being considered for inclusion in the next version of the EPPO diagnostic protocol?	yes
Kit	

Is a kit used	no
Other information	
Reaction type	Multiplex (>3)
Other details on the test	Quadruplex real-time PCR
Performance Criteria :	
Organism 1.:	Xanthomonas hortorum pv. gardneri(XANTGA)
Organism 2.:	Xanthomonas euvesicatoria pv. perforans(XANTPF)
Organism 3.:	Xanthomonas vesicatoria(XANTVE)
Organism 4.:	Xanthomonas euvesicatoria pv. euvesicatoria(XANTEU)
Analytical sensitivity	
What is smallest amount of target that can be detected reliably?	10 ⁴ cfu/mL (evaluated on tomato seed extracts spiked with bacterial suspensions at known concentration) 100 fg/ul (evaluated on bacterial DNA)
Diagnostic sensitivity	
Proportion of infected/infested samples tested positive compared to results from the standard test, see appendix 2 of PM 7/98	92%
Standard test(s)	Comparison of samples with known status
Analytical specificity - exclusivity	
Number of non-target organisms tested	Among 30 non-target strains false positive occurred on: CREA-DC 1032 Xanthomonas campestris pv. campestris, CREA-DC 1033 Xanthomonas hortorum pv. pelargonii, CREA-DC 1264 Xanthomonas citri pv. citri, NCPPB 1832 Xanthomonas arboricola pv. populi, NCPPB 1946 Xanthomonas campestris pv. raphani, CREA-DC 1495 unknown saprophyte strain from tomato seeds, LMG849 Xanthomonas axonopodis pv. poinsetticola.
Specificity value	76,6%
Diagnostic Specificity	
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	73%
Specify the test(s)	Comparison of samples with known status
Reproducibility	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	72%
Repeatability	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	75%

Test performance study	
Test performance study?	yes
Brief details of the test performance study and its output.It available, link to published article/report	Test performance study organized in the frame of a National Project (ASPROPI) involving seven Italian laboratories
The following complementary files are available online:	• VALIDATION_Xanthomonas_spp_ASPROPI_2017

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