

**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.

<b>Laboratory contact details</b>	Council for Agricultural Research and Economics– Research Centre for Plant Protection and Certification Via Carlo Giuseppe Bertero, 22, 00156 Rome, Italy
<b>Short description of the test</b>	detection of <i>Xanthomonas euvesicatoria</i> pv. <i>euvesicatoria</i> , <i>Xanthomonas hortorum</i> pv. <i>gardneri</i> , <i>Xanthomonas euvesicatoria</i> pv. <i>perforans</i> , <i>Xanthomonas vesicatoria</i> <i>Xanthomonas vesicatoria</i> by Molecular Conventional PCR in Seeds
<b>Date, reference of the validation report</b>	2022-01-22 - Validation_ASPROPI_2017_pp143-166
<b>Link to other validation data</b>	- Validation_ASPROPI_2017_pp143-166 detection of <i>Xanthomonas hortorum</i> pv. <i>gardneri</i> , <i>Xanthomonas euvesicatoria</i> pv. <i>perforans</i> , <i>Xanthomonas vesicatoria</i> , <i>Xanthomonas euvesicatoria</i> pv. <i>euvesicatoria</i> <i>Xanthomonas euvesicatoria</i> pv. <i>euvesicatoria</i> by Molecular real time RT PCR in Seeds
<b>Validation process according to EPPO Standard PM7/98?</b>	yes
<b>Is the lab accredited for this test?</b>	no
<b>Was the validated data generated in the framework of a project?</b>	Other_project
<b>If yes, please specify</b>	ASPROPI (Founded by MIPAAF_Italy)
<b>Description of the test</b>	
<b>Organism(s)</b>	<i>Xanthomonas euvesicatoria</i> pv. <i>euvesicatoria</i> (XANTEU) <i>Xanthomonas vesicatoria</i> (XANTVE) <i>Xanthomonas hortorum</i> pv. <i>gardneri</i> (XANTGA) <i>Xanthomonas euvesicatoria</i> pv. <i>perforans</i> (XANTPF)
<b>Detection / identification</b>	detection
<b>Matrix(ces) tested</b>	Seeds
<b>Plant species tested</b>	<i>Solanum lycopersicum</i>
<b>Method(s)</b>	Molecular Conventional PCR
<b>Method: Molecular Conventional PCR</b>	
<b>Reference of the test description</b>	
<b>As or adapted from an EPPO diagnostic</b>	yes

<b>protocol</b>	
<b>EPPO Diagnostic Protocol name</b>	PM 7/110 <i>Xanthomonas</i> spp. ( <i>Xanthomonas euvesicatoria</i> , <i>Xanthomonas gardneri</i> , <i>Xanthomonas perforans</i> , <i>Xanthomonas vesicatoria</i> ) causing bacterial spot of tomato and sweet pepper (version 1)
<b>Name of the test</b>	Conventional PCR (according to Koenraadt et al., 2009)
<b>As or adapted from an IPPC diagnostic protocol</b>	no
<b>Is the test modified compared to the reference test</b>	no
<b>Kit</b>	
<b>Is a kit used</b>	no
<b>Other information</b>	
<b>Reaction type</b>	Duplex
<b>Performance Criteria :</b>	
<b>Organism 1.:</b>	<b>Xanthomonas euvesicatoria pv. euvesicatoria(XANTEU)</b>
<b>Analytical sensitivity</b>	
<b>What is the smallest amount of target that can be detected reliably?</b>	10 <sup>4</sup> CFU/mL
<b>Diagnostic sensitivity</b>	
<b>Proportion of infected/infested samples tested positive compared to results from the standard test, see appendix 2 of PM 7/98</b>	95%
<b>Standard test(s)</b>	Comparison with samples of known status
<b>Analytical specificity - exclusivity</b>	
<b>Number of non-target organisms tested</b>	Among 26 non-target strains false positive occurred on: <i>Xanthomonas arboricola</i> pv. <i>celebensis</i> (NCPBP 1832), <i>Pseudomonas fluorescens</i> (NCPBP 1964), one unknown bacterial saprophyte isolate from tomato seeds (CREA-DC 1495).
<b>Specificity value</b>	88,4%
<b>Diagnostic Specificity</b>	
<b>Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test</b>	74%
<b>Specify the test(s)</b>	Comparison with samples of known status
<b>Reproducibility</b>	
<b>Provide the calculated % of agreement for a given level of the pest (see PM 7/98)</b>	74%
<b>Repeatability</b>	
<b>Provide the calculated % of agreement for a</b>	79%

given level of the pest (see PM 7/98)	
Organism 2.:	<b>Xanthomonas vesicatoria(XANTVE)</b>
<b>Analytical sensitivity</b>	
What is the smallest amount of target that can be detected reliably?	10 <sup>4</sup> CFU/mL
<b>Diagnostic sensitivity</b>	
Proportion of infected/infested samples tested positive compared to results from the standard test, see appendix 2 of PM 7/98	95%
Standard test(s)	Comparison with samples of known status
<b>Analytical specificity - exclusivity</b>	
Number of non-target organisms tested	Among 26 non-target strains false positive occurred on: Xanthomonas arboricola pv. celebensis (NCPBP 1832), Pseudomonas fluorescens (NCPBP 1964), one unknown bacterial saprophyte isolate from tomato seeds (CREA-DC 1495).
Specificity value	88,4%
<b>Diagnostic Specificity</b>	
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	74%
Specify the test(s)	Comparison with samples of known status
<b>Reproducibility</b>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	74%
<b>Repeatability</b>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	79%
Organism 3.:	<b>Xanthomonas hortorum pv. gardneri(XANTGA)</b>
<b>Analytical sensitivity</b>	
What is the smallest amount of target that can be detected reliably?	10 <sup>4</sup> CFU/mL
<b>Diagnostic sensitivity</b>	
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
<b>Analytical specificity - exclusivity</b>	
Number of non-target organisms tested	Among 26 non-target strains false positive occurred on: Xanthomonas arboricola pv. celebensis (NCPBP 1832), Pseudomonas fluorescens (NCPBP 1964), one unknown bacterial saprophyte isolate from tomato seeds (CREA-DC 1495).

<b>Specificity value</b>	88,4%
<b><u>Diagnostic Specificity</u></b>	
<b>Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test</b>	82%
<b>Specify the test(s)</b>	Comparison of samples with known status
<b><u>Reproducibility</u></b>	
<b>Provide the calculated % of agreement for a given level of the pest (see PM 7/98)</b>	78%
<b><u>Repeatability</u></b>	
<b>Provide the calculated % of agreement for a given level of the pest (see PM 7/98)</b>	81%
<b>Organism 4.:</b>	<b>Xanthomonas euvesicatoria pv. perforans(XANTPF)</b>
<b><u>Analytical sensitivity</u></b>	
<b>What is the smallest amount of target that can be detected reliably?</b>	10 <sup>4</sup> CFU/mL
<b><u>Diagnostic sensitivity</u></b>	
<b>Proportion of infected/infested samples tested positive compared to results from the standard test, see appendix 2 of PM 7/98</b>	92%
<b>Standard test(s)</b>	Comparison with samples of known status
<b><u>Analytical specificity - exclusivity</u></b>	
<b>Number of non-target organisms tested</b>	Among 26 non-target strains false positive occurred on: Xanthomonas arboricola pv. celebensis (NCPBP 1832), Pseudomonas fluorescens (NCPBP 1964), one unknown bacterial saprophyte isolate from tomato seeds (CREA-DC 1495).
<b>Specificity value</b>	88,4%
<b><u>Diagnostic Specificity</u></b>	
<b>Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test</b>	82%
<b>Specify the test(s)</b>	Comparison with samples of known status
<b><u>Reproducibility</u></b>	
<b>Provide the calculated % of agreement for a given level of the pest (see PM 7/98)</b>	78%
<b><u>Repeatability</u></b>	
<b>Provide the calculated % of agreement for a given level of the pest (see PM 7/98)</b>	81%
<b><u>Test performance study</u></b>	
<b>Test performance study?</b>	no
<b>Brief details of the test performance study</b>	Test performance study organized in the frame of a

<b>and its output.It available, link to published article/report</b>	National Project (ASPROPI) involving 7 Italian laboratories
The following complementary files are available online:	<ul style="list-style-type: none"><li>• <a href="#">VALIDATION_Xanthomonas_spp_ASPROPI_2017</a></li></ul>

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