

**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>	Molecular detection of <i>Xanthomonas citri</i> pv. <i>citri</i> (lemon leaves and orange fruits) and <i>Xanthomonas citri</i> pv. <i>aurantifolii</i> (lime fruits) by cPCR and real-time PCR
<b>Date, reference of the validation report</b>	2022-09-28 - Report_EURL-PT-2021-01-Xc_IV
<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>	EURL
<b>Description of the test</b>	
<b>Organism(s)</b>	<i>Xanthomonas citri</i> pv. <i>aurantifolii</i> (XANTAU) <i>Xanthomonas citri</i> pv. <i>citri</i> (XANTCI)
<b>Detection / identification</b>	detection
<b>Matrix(ces) tested</b>	Fruits, Leaves
<b>Plant species tested</b>	<i>Citrus x aurantiifolia</i> , <i>Citrus x aurantium</i> var. <i>sinensis</i> , <i>Citrus x limon</i>
<b>Method(s)</b>	Molecular Conventional PCR Molecular Conventional PCR (2) Molecular real time PCR Molecular real time PCR (2) Molecular real time PCR (3)
<b>Method: Molecular Conventional PCR</b>	
<b>Reference of the test description</b>	
<b>As or adapted from an EPPO diagnostic protocol</b>	no
<b>New test being considered for inclusion in the next version of the EPPO diagnostic protocol?</b>	yes
<b>As or adapted from an IPPC diagnostic protocol</b>	no
<b>Reference of the test</b>	Mavrodieva et al., 2004

<b>Kit</b>	
<b>Is a kit used</b>	no
<b>Other information</b>	
<b>Reaction type</b>	Simplex
<b>Other details on the test</b>	The targets of this test are both <i>Xanthomonas citri</i> pv. <i>citri</i> and <i>Xanthomonas citri</i> pv. <i>aurantifolii</i> .
<b>Method: Molecular Conventional PCR (2)</b>	
<b>Reference of the test description</b>	
<b>As or adapted from an EPPO diagnostic protocol</b>	no
<b>New test being considered for inclusion in the next version of the EPPO diagnostic protocol?</b>	yes
<b>As or adapted from an IPPC diagnostic protocol</b>	no
<b>Reference of the test</b>	Robène et al., 2020
<b>Kit</b>	
<b>Is a kit used</b>	no
<b>Other information</b>	
<b>Reaction type</b>	Simplex
<b>Other details on the test</b>	The target of this test is <i>Xanthomonas citri</i> pv. <i>citri</i> .
<b>Method: Molecular real time PCR</b>	
<b>Reference of the test description</b>	
<b>As or adapted from an EPPO diagnostic protocol</b>	no
<b>New test being considered for inclusion in the next version of the EPPO diagnostic protocol?</b>	yes
<b>As or adapted from an IPPC diagnostic protocol</b>	yes
<b>IPPC diagnostic Protocol name</b>	(version )
<b>Name of the test</b>	Cubero and Graham, 2005
<b>Kit</b>	
<b>Is a kit used</b>	no
<b>Other information</b>	
<b>Reaction type</b>	Simplex - Probe
<b>Other details on the test</b>	TaqMan. The target of this test is <i>Xanthomonas citri</i> pv. <i>citri</i> .
<b>Method: Molecular real time PCR (2)</b>	
<b>Reference of the test description</b>	
<b>As or adapted from an EPPO diagnostic protocol</b>	no

<b>New test being considered for inclusion in the next version of the EPPO diagnostic protocol?</b>	yes
<b>As or adapted from an IPPC diagnostic protocol</b>	no
<b>Reference of the test</b>	Mavrodieva et al., 2004
<b>Kit</b>	
<b>Is a kit used</b>	no
<b>Other information</b>	
<b>Reaction type</b>	Simplex
<b>Other details on the test</b>	SYBR Green. The targets of this test are both <i>Xanthomonas citri</i> pv. <i>citri</i> and <i>Xanthomonas citri</i> pv. <i>aurantifolii</i> .
<b>Method: Molecular real time PCR (3)</b>	
<b>Reference of the test description</b>	
<b>As or adapted from an EPPO diagnostic protocol</b>	no
<b>New test being considered for inclusion in the next version of the EPPO diagnostic protocol?</b>	yes
<b>As or adapted from an IPPC diagnostic protocol</b>	no
<b>Reference of the test</b>	Robène et al., 2020
<b>Kit</b>	
<b>Is a kit used</b>	no
<b>Other information</b>	
<b>Reaction type</b>	Duplex - Probe
<b>Other details on the test</b>	TaqMan. The target of this test is <i>Xanthomonas citri</i> pv. <i>citri</i> .
<b>Performance Criteria :</b>	
<b>Organism 1.:</b>	<b><i>Xanthomonas citri</i> pv. <i>aurantifolii</i>(XANTAU)</b>
<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>	- cPCR (Mavrodieva et al., 2004): 100%; - real-time PCR (Mavrodieva et al., 2004): 99%.
<b>Standard test(s)</b>	Comparison with samples of known status.
<b>Diagnostic Specificity</b>	
<b>Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test</b>	- cPCR (Mavrodieva et al., 2004): 98%; - real-time PCR (Mavrodieva et al., 2004): 96%.
<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>	- cPCR (Mavrodieva et al., 2004): 99%; - real-time PCR (Mavrodieva et al., 2004): 98%.

<b>Organism 2.:</b>	<b>Xanthomonas citri pv. citri(XANTCI)</b>
<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>	- cPCR (Robène et al., 2020): 100% - real-time PCR (Robène et al., 2020): 100% - real-time PCR (Cubero and Graham, 2005): 98%
<b>Standard test(s)</b>	Comparison with samples of known status.
<b>Diagnostic Specificity</b>	
<b>Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test</b>	- cPCR (Robène et al., 2020): 100% - real-time PCR (Robène et al., 2020): 86% - real-time PCR (Cubero and Graham, 2005): 81%
<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>	- cPCR (Robène et al., 2020): 100% - real-time PCR (Robène et al., 2020): 94% - real-time PCR (Cubero and Graham, 2005): 89%
<b>Test performance study</b>	
<b>Test performance study?</b>	no
<b>Brief details of the test performance study and its output.It available, link to published article/report</b>	Proficiency test organised in the framework of EURL activity. This activity allowed to obtain informations on the proficiency of tests used by the participants. Number of participants for each test: - cPCR Robène et al., 2020: 17 participants; - cPCR Mavrodieva et al., 2004: 14 participants; - real-time PCR Mavrodieva et al., 2004: 16 participants; - real-time PCR Robène et al., 2020: 15 participants; - real-time PCR Cubero and Graham, 2005: 14 participants;
<b>Other information</b>	
<b>Any other information considered useful</b>	Each participant received a panel samples represented by: DNA extracts of three plant matrices added with bacterial DNA at known concentration following the scheme reported in the attached file (see test items panel).
The following complementary files are available online:	
	<ul style="list-style-type: none"> <li>• <a href="#">EURL_presentation Xcc_Xca</a></li> </ul>

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