

**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>	Anses Plant Health Laboratory - Pests and Tropical Pathogens Unit Pôle de Protection des Plantes, 7 Chemin de l'IRAT, 97410 Saint Pierre, France
<b>Short description of the test</b>	Detection and identification of 'Candidatus Liberibacter asiaticus' and 'Candidatus Liberibacter africanus' by Molecular duplex conventional PCR in Citrus sp. leaves
<b>Date, reference of the validation report</b>	2020-07-10 - HLB_qPCR_EUPHRESCO-2016-A-232
<b>Link to other validation data</b>	- HLB_qPCR_EUPHRESCO-2016-A-232 Detection and identification of 'Candidatus Liberibacter asiaticus' and 'Candidatus Liberibacter africanus' by Molecular duplex conventional PCR in Citrus sp. leaves
<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>	Euphresco
<b>If yes, please specify</b>	2016-A-232
<b>Description of the test</b>	
<b>Organism(s)</b>	'Candidatus Liberibacter asiaticus'(LIBEAS) 'Candidatus Liberibacter africanus'(LIBEAF)
<b>Detection / identification</b>	detection and identification
<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 protocol</b>	no
<b>As or adapted from an IPPC diagnostic protocol</b>	no
<b>Reference of the test</b>	Conventional PCR targeting 16S rRNA gene (according to Teixeira et al., 2005a,b) duplexed with the conventional PCR targeting rplKAJL - rpoBC operon gene (according to Hocquellet et al., 1999)

Is the test modified compared to the reference test	yes Two conventional PCR were duplexed
<b>Kit</b>	
Is a kit used	no
<b>Other information</b>	
Reaction type	Duplex
Are the performance characteristics included in the EPP0 diagnostic protocol?	no
<b>Performance Criteria :</b>	
Organism 1.:	' <b>Candidatus Liberibacter asiaticus</b> '(LIBEAS)
<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	0.868 (DSE=PA/N+, Fav. Hypothesis, considering CLas and CLaf samples)
<b>Diagnostic Specificity</b>	
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	1 (DSP=NA/N-, Fav. Hypothesis, considering CLas and CLaf samples)
<b>Reproducibility</b>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	CO=0.952 (considering CLas and CLaf samples)
<b>Repeatability</b>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	DA=1 (considering CLas and CLaf samples)
Organism 2.:	' <b>Candidatus Liberibacter africanus</b> '(LIBEAF)
<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	0.868 (DSE=PA/N+, Fav. Hypothesis, considering CLas and CLaf samples)
<b>Diagnostic Specificity</b>	
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	1 (DSP=NA/N-, Fav. Hypothesis, considering CLas and CLaf samples)
<b>Reproducibility</b>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	CO=0.952 (considering CLas and CLaf samples)
<b>Repeatability</b>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	DA=1 (considering CLas and CLaf samples)
<b>Test performance study</b>	
Test performance study?	yes
Brief details of the test performance study	Test performance study organized in the

<b>and its output.It available, link to published article/report</b>	framework of a EUPHRESKO project involving 8 international laboratories.
<b>Other information</b>	
<b>Any other information considered useful</b>	Publication available at: <a href="https://link.springer.com/content/pdf/10.1007/s10658-020-02052-3.pdf">https://link.springer.com/content/pdf/10.1007/s10658-020-02052-3.pdf</a> Cellier, G., C. Redondo, J. Cubero, M. Roselló, E. de Andrade, L. Cruz, E. Ince, H. N. Yildiz, P. G. Güler, A. M. D'Onghia, T. Yaseen, K. Djelouah, E. Metz-Verschure, F. Gaffuri, R. A. Gottsberger, and B. Giovani. 2020. "Comparison of the performance of the main real-time and conventional PCR detection tests for 'Candidatus Liberibacter' spp., plant pathogenic bacteria causing the Huanglongbing disease in Citrus spp." European Journal of Plant Pathology. doi: 10.1007/s10658-020-02052-3.

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