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	Anses Plant Health Laboratory - Pests and Tropical Pathogens Unit Pôle de Protection des Plantes, 7 Chemin de l'IRAT, 97410 Saint Pierre, France
Short description of the test	Detection and identification of 'Candidatus Liberibacter asiaticus' and 'Candidatus Liberibacter africanus' by Molecular duplex conventional PCR in Citrus sp. leaves
Date, reference of the validation report	2020-07-10 - HLB_qPCR_EUPHRESCO-2016-A-232
Link to other validation data	- 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
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?	Euphresco
If yes, please specify	2016-A-232

Description of the test	
Organism(s)	'Candidatus Liberibacter asiaticus' (LIBEAS) 'Candidatus Liberibacter africanus' (LIBEAF)
Detection / identification	detection and identification
Method(s)	Molecular Conventional PCR
Method: Molecular Conventional PCR	
Reference of the test description	
As or adapted from an EPPO diagnostic protocol	no
As or adapted from an IPPC diagnostic protocol	no
Reference of the test	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
Kit	
Is a kit used	no
Other information	
Reaction type	Duplex
Are the performance characteristics included in the EPPO diagnostic protocol?	**no**
Performance Criteria :	
Organism 1.:	**'Candidatus Liberibacter asiaticus'(LIBEAS)**
<u>Diagnostic sensitivity</u>	
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)
<u>Diagnostic Specificity</u>	
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)
<u>Reproducibility</u>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	CO=0.952 (considering CLas and CLaf samples)
<u>Repeatability</u>	
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.:	**'Candidatus Liberibacter africanus'(LIBEAF)**
<u>Diagnostic sensitivity</u>	
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)
<u>Diagnostic Specificity</u>	
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)
<u>Reproducibility</u>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	CO=0.952 (considering CLas and CLaf samples)
<u>Repeatability</u>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	DA=1 (considering CLas and CLaf samples)
Test performance study	
Test performance study?	yes
Brief details of the test performance study	Test performance study organized in the

and its output.It available, link to published article/report	framework of a EUPHRESCO project involving 8 international laboratories.
Other information	
Any other information considered useful	Publication available at: https://link.springer.com/content/pdf/10.1007/s10658-020-02052-3.pdf 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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