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	Bacteriology. Instituto Valenciano de Investigaciones Agrarias CV-315, km. 10.7, 46113 Moncada, Spain
Short description of the test	Detection of 'Candidatus Liberibacter solanacearum' by real time PCR in carrot seeds using Plant Print diagnòstics kit
Date, reference of the validation report	2016-05-02 - Report 2016/05/02; Validation assay June 2015 - PNT-18/2015
Validation process according to EPPO Standard PM7/98?	yes
Is the lab accredited for this test?	yes
Was the validated data generated in the framework of a project?	no
Description of the test	
Organism(s)	'Candidatus Liberibacter solanacearum' (LIBEPS)
Detection / identification	detection
Method(s)	Extraction Molecular real time PCR
Method: Extraction	
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	Bertolini et al. 2014a, Teresani et al. 2014
Other information	
Other details on the test	Direct sample preparation without DNA purification (spot procedure) (Bertolini et al. 2014a, Teresani et al. 2014)
Method: Molecular real time PCR	
Reference of the test description	
As or adapted from an EPPO diagnostic protocol	yes

EPPO Diagnostic Protocol name	PM 7/143 ‘Candidatus Liberibacter solanacearum’ (version 1)
Name of the test	Real-time PCR based on 16S rRNA gene (Teresani et al., 2014)
Is the test modified compared to the reference test	yes Use of a kit
Kit	
Is a kit used	yes
Manufacturer name	PLANT PRINT
Specify the kit used	'Candidatus Liberibacter solanacearum' Complete real-time PCR kit for direct screening (Ref: CaLsol/100)
Kit used following the manufacturer's instructions?	
Other information	
Other details on the test	Real time PCR using Plant Print diagnostic kit, based on Bertolini et al. 2014
Are the performance characteristics included in the EPPO diagnostic protocol?	**no**
Performance Criteria :	
Organism 1.:	**'Candidatus Liberibacter solanacearum'(LIBEPS)**
Analytical sensitivity	
What is smallest amount of target that can be detected reliably?	Not calculated for a non-culturable bacterium. The performance study was oriented to receive qualitative results.
Diagnostic sensitivity	
Proportion of infected/infested samples tested positive compared to results from the standard test, see appendix 2 of PM 7/98	100% (Standard test was real time PCR according to Bertolini et al. after CTAB extraction)
Standard test(s)	75 samples agreement / 75 (including replications performed in some labs)
Diagnostic Specificity	
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	100% (Standard test was real time PCR according to Bertolini et al. after CTAB extraction)
Specify the test(s)	75 samples agreement / 75 (including replications performed in some labs)
Reproducibility	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100% (150/150)
Repeatability	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100% The repeatability was calculated in 5 laboratories that performed 2 replications

Test performance study	
Test performance study?	yes
Brief details of the test performance study and its output.It available, link to published article/report	Ring test during acreditation process. 10 official Laboratories of Diagnostic of Spain tested this method: Laboratorio de Producción y Sanidad Vegetal, Huelva; Laboratorio de Producción y Sanidad Vegetal, Sevilla; Laboratorio de Sanidad Vegetal-ICIA, Tenerife; Centro Regional de Diagnóstico, Salamanca; Laboratorio de Diagnóstico Fitopatológico (Bacteriología), Valencia; Laboratorio de Bacteriologia- IVIA, Valencia; Laboratorio Nacional de Referencia de Bacteriología (MAGRAMA), Valencia; Laboratorio Regional de la CC. AA. de La Rioja, Logroño; Laboratorio de Bacteriologia-INIA, Madrid; Sanidad Vegetal-INIA, Madrid. The test performance study was organized by IVIA.
Other information	
Any other information considered useful	The diagnostic kit evaluated is simple to use, rapid and accurate. It showed a high robustness in 10 laboratories, and can be applied for rapid testing of carrot seeds. For maximum accuracy a previous CTAB extraction or other types of DNA extraction is adviced.
The following complementary files are available online:	• Ejercicio colaborativo CaLsol

Creation date: 2016-05-02 00:00:00 - Last update: 2020-10-27 17:41:30