EUROPEAN AND MEDITERRANEAN PLANT PROTECTION ORGANIZATION ORGANISATION EUROPEENNE ET MEDITERRANEENNE POUR LA PROTECTION DES PLANTES (11-17239)

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.

Target Organism	Candidatu	us Liberibacter solanacearum
Short description	Detection of 'Candidatus Liberibacter solanacearum' by means of the real-time PCR procedure	
Laboratory contact details	ILVO Institute for Agricultural and Fisheries Research Burg. Van Gansberghelaan 96, 9820 Merelbeke - Melle, Belgium	
Date and reference of the validation report	28/06/2016 - F16_V13	
Validation process according to EPPO Standard PM 7/98:	Yes	
Reference of the test description	0 DP21 (IPPC) Li et al. 2009 Li W., Abad J.A., French-Monar R.D., Rascoe J., Wen A., Gudmestad N.C., Secor G.A., Lee I-M., Duan Y., Levy L. Multiplex real-time PCR for detection, identification and quantification of 'Candi-datus Liberibacter solanacearum' in potato plants with zebra chip. Journal of Microbiological Meth-ods 78, 59–65 (2009).	
Is the test the same as described in the EPPO DP?	Yes	
Is the lab accredited for this test?	Yes	
Plant species tested (if relevant)	Solanum tuberosum (potato) and Daucus carota (carrot)	
Matrices tested (if relevant)	leaves, petioles, roots	
List of methods used	1	
Method for extraction / isolation / baiting of target organism from matrix	X	DNeasy (Qiagen)
Molecular methods, e.g. hybridization, PCR and real time PCR	X	DP21 (IPPC) Li et al. 2009
Serological methods: IF, ELISA, Direct Tissue Blot Immuno Assay		
Plating methods: selective isolation		
Bioassay methods: selective enrichment in host plants, baiting, plant test and grafting.		
Pathogenicity test		

Fingerprint methods: protein profiling, fatty acid profiling & DNA profiling		
Morphological and morphometrical methods intended for identification		
Biochemical methods: e.g. enzyme electrophoresis, protein profiling		
Other		
Analytical sensitivity (= limit of detection	<u>ction)</u>	
What is smallest amount of target that can be detected reliably?		
Diagnostic sensitivity		
Proportion of infected/infested samples tested positive compared to results from the standard test , see appendix 2 of PM 7/98		
Specify the standard test		
Analytical specificity		
Specificity value		
Number of strains/populations of target organisms tested	2 Ca. L. solanacearum Isolate potato (USDA, Venkat, Texas, USA) RefV_CaLso_01 Ca. L. solanacearum Isolate carrot (origin Marocco) RefV_CaLso_02	
Number of non-target organisms tested	20 Arabis mosaic virus Cucumber mosaic virus Potato leaf roll virus Potato virus Y Potato virus X Potato spindle tuber viroid Strawberry latent ringspot virus Tobacco rattle virus Tomato black ring virus Tomato black ring virus Clavibacter michiganensis subsp. sepedonicus Dickeya solani Ralstonia solanacearum Rhizoctonia solani Verticillium dahliae Candidatus Phytoplasma asteris (aster yellows fytoplasma) Candidatus Phytoplasma solani (stolbur fytoplasma) Candidatus Liberibacter asiaticus Candidatus Liberibacter africanus Candidatus Liberibacter americanus	
Cross reacts with (specify the	In some cases, late reaction (Cq >38) for Ca L americanus	

species)	observed	
Diagnostic Specificity		
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test		
Specify the standard test		
Reproducibility		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100%	
Repeatability		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100%	
Test performance study		
Test performance study?	Yes	
Include brief details of the test performance study and its output.It available, provide a link to published article/report	2017 - ANSES (Test performance study, including two real- time PCR methods (Li et al 2009 and Teresani et al 2014) and three conventional PCR (Li et al 2009; Munyaneza et al 2009, Ravindran et al 2011)	
Other information		
Any other information considered useful e.g. robustness, ease of performing the test, etc.		