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	Clavibacter sepedonicus	
Short description	Detection of Clavibacter sepedonicus in potato extract by conventional PCR, in a multiplex assay with Ralstonia solanacearum	
Laboratory contact details	Bavarian State Research Center for Agriculture, Institute for Plant Protection - Phytopathology and Diagnosis Lange Point 10, 85354 Freising, Germany	
Date and reference of the validation report	2018-04-16 - n/a	
Validation process according to EPPO Standard PM 7/98:	Yes	
Reference of the test description	PM 7/059 Pastrik K-H (2000) Detection of Clavibacter michiganensis ssp. sepedonicus in potato tubers by multiplex PCR with coamplification of host DNA. Eur. J. Plant Pathol. 106, 155-165.	
Is the test the same as described in the EPPO DP?	Modified - PCR mastermix (Qiagen Multiplex PCR Plus Kit) - DNA extraction: MasterPure Complete DNA Purification kit (Lucigen) - Multiplex setup with primers Rs 1 F/R for Rs (Pastrik et al., 2002) - IPC after White et al., 1990 (primer NS7, NS8)	
Is the lab accredited for this test?	Yes	
Plant species tested (if relevant)	Solanum tuberosum	
Matrices tested (if relevant)	tuber extract	
List of methods used		
Method for extraction / isolation / baiting of target organism from matrix		
Molecular methods, e.g. hybridization, PCR and real time PCR	X	conventional PCR
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)		
What is smallest amount of target that can be detected reliably?	10^3 cells/ ml tuber extract	
Diagnostic sensitivity		
Proportion of infected/infested samples tested positive compared to results from the standard test , see appendix 2 of PM 7/98	not done	
Specify the standard test	n/a	
Analytical specificity		
Specificity value	100%	
Number of strains/populations of target organisms tested	6 LMG 2894 LMG 2889 NCPPB 3898 LMG 6722 NCPPB 2140 LMG 25595	
Number of non-target organisms tested	10 Clavibacter michiganensis subsp. michiganensis LMG 3687 Clavibacter michiganensis subsp. insidiosus LMG 7268 Pseudomonas syringae pv. striafaciens GSPB 2570 Pectobacterium atrosepticum SCRI 1039 Pectobacterium carotovorum subsp. carotovorum LMG 2401 Pectobacterium wasabiae DSM 18074 Pectobacterium carotovorum subsp. brasiliensis LMG 21371 Pectobacterium carotovorum subsp. odoriferum LMG 6688 Pectobacterium betavasculorum LMG 2466 Dickeya solani JKI	
Cross reacts with (specify the species)	none	
Diagnostic Specificity		
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	not done	

Specify the standard test	n/a
Reproducibility	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100% for 10^3 cells/ ml (a total of 56 PCR reactions containing 10^3 samples (in 24 PCR runs, two different operators, different days), of which 56 were positive)
Repeatability	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100% for 10^3 cells/ ml (a total of 28 PCR reactions containing 10^3 samples, in 12 PCR runs, each repeated once - same day, same operator -, of which 28 with the same result)
Test performance study	
Test performance study?	No
Include brief details of the test performance study and its output.It available, provide a link to published article/report	
Other information	
Any other information considered useful e.g. robustness, ease of performing the test, etc.	