

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	Tomato chlorosis virus	
Short description	Detection of ToCV by RT-PCR in tomato leaves	
Laboratory contact details	Anses Plant Health Laboratory - Bacteriology, Virology and GMO Unit 7 rue Jean Dixm�ras, 49000 Angers, France	
Date and reference of the validation report	2011-07 - Loiseau M. et Cousseau P. 2011. Evaluation des m�thodes de d�tection des jaunisses de la tomate - Tomato Infectious Chlorosis Virus (TICV) Tomato Chlorosis Virus (ToCV)	
Validation process according to EPPO Standard PM 7/98:	No	
Reference of the test description	0 Jacquemond M., Verdin E., Dalmon A., Guilbaud L., Gognalons P., 2009. Serological and molecular detection of Tomato chlorosis virus and Tomato infectious chlorosis virus in tomato. <i>Plant pathology</i> 58:210-220. Louro D., Accotto G.P., Vaira A.M., 2000. Occurrence and diagnosis of Tomato chlorosis virus in Portugal. <i>European Journal of Plant Pathology</i> , 106: 589-592	
Is the test the same as described in the EPPO DP?	No -	
Is the lab accredited for this test?	No	
Plant species tested (if relevant)	Solanum esculentum	
Matrices tested (if relevant)	leaves	
List of methods used		
Method for extraction / isolation / baiting of target organism from matrix	X	For the RNA extraction, leaf samples was grinded in the RLT buffer (quiagen)
Molecular methods, e.g. hybridization, PCR and real time PCR	X	RNA was extracted with the Plant RNeasy minikit from Qiagen. RT-PCR tests were carried out following the recommendation of the paper of Jacquemond et al (2009) et Louro et al (2000).
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		
<u>Analytical sensitivity (= limit of detection)</u>		
What is smallest amount of target that can be detected reliably?	Not relevant	
<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	Simplex RT-PCR (Louro, 2000): 86.67% to 88.89%; Duplex RT-PCR (Jacquemon, 2009): 83.3%; Triplex Rt-PCR (with Cox) (Jacquemon, 2009): 49.02% to 54.9%	
Specify the standard test		
<u>Analytical specificity</u>		
Specificity value		
Number of strains/populations of target organisms tested	15 (see table as separate file or full validation report for detail)	
Number of non-target organisms tested	22 (see table as separate file or full validation report for detail)	
Cross reacts with (specify the species)	Cross reaction observed with the triplex method	
<u>Diagnostic Specificity</u>		
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	Simplex RT-PCR (Louro, 2000): 100%; Duplex RT-PCR (Jacquemon, 2009): 100%; Triplex Rt-PCR (with Cox) (Jacquemon, 2009): 97.5% to 100%	
Specify the standard test		
<u>Reproducibility</u>		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)		
<u>Repeatability</u>		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)		
<u>Test performance study</u>		
Test performance study?	No	

<p>Include brief details of the test performance study and its output. If available, provide a link to published article/report</p>	
<p><u>Other information</u></p>	
<p>Any other information considered useful e.g. robustness, ease of performing the test, etc.</p>	
<p>The following complementary files are available online:</p>	<ul style="list-style-type: none"> • List of target strains and non-target organisms • Loiseau M. et Cousseau P. 2011. Evaluation des méthodes de détection des jaunisses de la tomate – Tomato Infectious Chlorosis Virus (TICV) Tomato Chlorosis Virus (ToCV)