## 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 cl	nlorosis 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 Dixméras, 49044 Angers, France	
Date and reference of the validation report	2011-07 - 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)	
Validation process according to EPPO Standard PM 7/98:	No	
Reference of the test description	0 Jacquemo P., 2009. chlorosis tomato. P Vaira A.M chlorosis Pathology	nd M., Verdin E., Dalmon A., Guilbaud L., Gognalons Serological and molecular detection of Tomato virus and Tomato infectious chlorosis virus in lant pathology 58:210-220. Louro D., Accotto G.P., ., 2000. Occurrence and diagnosis of Tomato virus in Portugal. European Journal of Plant 7, 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 e	esculentum
Matrices tested (if relevant)	leaves	
List of methods used		
Method for extraction / isolation / baiting of target organism from matrix	Х	For the RNA extraction, leaf samples was grinded in the RLT buffer (quiagen)
Molecular methods, e.g. hybridization, PCR and real time PCR	Х	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				
Analytical sensitivity (= limit of detection)				
What is smallest amount of target that can be detected reliably?	Not relevant			
Diagnostic sensitivity				
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 (Jacquemond, 2009): 83.3%; Triplex Rt-PCR (with Cox) (Jacquemond, 2009): 49.02% to 54.9%			
Specify the standard test				
Analytical specificity				
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 ta	ble as separate file or full validation report for detai		
Number of non-target organisms tested Cross reacts with (specify the species)	22 (see ta Cross read	ble as separate file or full validation report for detai		
Number of non-target organisms testedCross reacts with (specify the species)Diagnostic Specificity	22 (see ta Cross read	ble as separate file or full validation report for detai		
Number of non-target organisms testedCross reacts with (specify the species)Diagnostic SpecificityProportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	22 (see ta Cross read Simplex R (Jacquemo	ble as separate file or full validation report for detai etion observed with the triplex method T-PCR (Louro, 2000): 100%; Duplex RT-PCR ond, 2009): 100%; Triplex Rt-PCR (with Cox) ond, 2009): 97.5% to 100%		
Number of non-target organisms testedCross reacts with (specify the species)Diagnostic SpecificityProportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard testSpecify the standard test	22 (see ta Cross read Simplex R (Jacquemo (Jacquemo	ble as separate file or full validation report for detai etion observed with the triplex method T-PCR (Louro, 2000): 100%; Duplex RT-PCR ond, 2009): 100%; Triplex Rt-PCR (with Cox) ond, 2009): 97.5% to 100%		
Number of non-target organisms testedCross reacts with (specify the species)Diagnostic SpecificityProportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard testSpecify the standard testReproducibility	22 (see ta Cross read Simplex R (Jacquemo (Jacquemo	ble as separate file or full validation report for detai etion observed with the triplex method T-PCR (Louro, 2000): 100%; Duplex RT-PCR ond, 2009): 100%; Triplex Rt-PCR (with Cox) ond, 2009): 97.5% to 100%		
Number of non-target organisms testedCross reacts with (specify the species)Diagnostic SpecificityProportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard testSpecify the standard testReproducibilityProvide the calculated % of agreement for a given level of the pest (see PM 7/98)	22 (see ta Cross read Simplex R (Jacquemo (Jacquemo	ble as separate file or full validation report for detai etion observed with the triplex method T-PCR (Louro, 2000): 100%; Duplex RT-PCR ond, 2009): 100%; Triplex Rt-PCR (with Cox) ond, 2009): 97.5% to 100%		
Number of non-target organisms testedCross reacts with (specify the species)Diagnostic SpecificityProportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard testSpecify the standard testSpecify the standard testReproducibilityProvide the calculated % of agreement for a given level of the pest (see PM 7/98)Repeatability	22 (see ta Cross read Simplex R (Jacquemo (Jacquemo	ble as separate file or full validation report for detai etion observed with the triplex method T-PCR (Louro, 2000): 100%; Duplex RT-PCR ond, 2009): 100%; Triplex Rt-PCR (with Cox) ond, 2009): 97.5% to 100%		
Number of non-target organisms testedCross reacts with (specify the species)Diagnostic SpecificityProportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard testSpecify the standard testSpecify the standard testReproducibilityProvide the calculated % of agreement for a given level of the pest (see PM 7/98)RepeatabilityProvide the calculated % of agreement for a given level of the pest (see PM 7/98)	22 (see ta Cross read Simplex R (Jacquemo (Jacquemo	ble as separate file or full validation report for detai etion observed with the triplex method T-PCR (Louro, 2000): 100%; Duplex RT-PCR ond, 2009): 100%; Triplex Rt-PCR (with Cox) ond, 2009): 97.5% to 100%		
Number of non-target organisms testedCross reacts with (specify the species)Diagnostic SpecificityProportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard testSpecify the standard testSpecify the standard testReproducibilityProvide the calculated % of agreement for a given level of the pest (see PM 7/98)RepeatabilityProvide the calculated % of agreement for a given level of the pest (see PM 7/98)Test performance study	22 (see ta Cross read Simplex R (Jacquemo (Jacquemo	ble as separate file or full validation report for detai ation observed with the triplex method T-PCR (Louro, 2000): 100%; Duplex RT-PCR and, 2009): 100%; Triplex Rt-PCR (with Cox) and, 2009): 97.5% to 100%		

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.	
The following complementary files are available online:	<ul> <li>List of target strains and non-target organisms</li> <li>Loiseau M. et Cousseau P. 2011. Evaluation des</li> </ul>
	méthodes de détection des jaunisses de la tomate – Tomato Infectious Chlorosis Virus (TICV) Tomato Chlorosis Virus (ToCV)