

**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.

<b>Laboratory contact details</b>	ILVO Institute for Agricultural and Fisheries Research Burg. Van Gansberghelaan 96, 9820 Merelbeke, Belgium
<b>Short description of the test</b>	Screening TAS-ELISA test for Tomato yellow leaf curl virus - Antibody set ADGEN/NEOGEN 1072-25
<b>Date, reference of the validation report</b>	2018-02-12 - F16_V01; F16_V06
<b>Validation process according to EPPO Standard PM7/98?</b>	yes
<b>Is the lab accredited for this test?</b>	yes
<b>Was the validated data generated in the framework of a project?</b>	
<b>Description of the test</b>	
<b>Organism(s)</b>	Begomovirus coheni(TYLCV0)
<b>Detection / identification</b>	detection
<b>Method(s)</b>	Molecular real time PCR Serological DASi-ELISA
<b>Method: Molecular real time PCR</b>	
<b>Reference of the test description</b>	
<b>Other information</b>	
<b>Other details on the test</b>	method of fera, UK; EUPHRESKO final report project "Validation of diagnostic methods for the detection and identification of whitefly transmitted viruses of regulatory or quarantine concern to the EU."
<b>Method: Serological DASi-ELISA</b>	
<b>Reference of the test description</b>	
<b>As or adapted from an EPPO diagnostic protocol</b>	yes
<b>EPPO Diagnostic Protocol name</b>	PM 7/050 Tomato yellow leaf curl and Tomato mottle begomoviruses (version 1)
<b>Name of the test</b>	TAS-ELISA (based on Thomas, 1986)
<b>Is the test modified compared to the reference test</b>	no

<b>Other information</b>	
<b>Other details on the test</b>	the ELISA is used as a screening test. Positive results are submitted to one-step real-time PCR procedure TAS-ELISA screening (AB set 1072-25, Adgen/neogen phytodiagnostics)
<b>Performance Criteria :</b>	
<b>Organism 1.:</b>	<b>Begomovirus coheni(TYLCV0)</b>
<b>Analytical sensitivity</b>	
<b>What is smallest amount of target that can be detected reliably?</b>	Because the concentration of viruses, viroids and phytoplasmas is never known, determine the maximum dilution of RNA /DNA detected. Therefore, the sensitivity determined here is not an absolute sensitivity but a relative sensitivity.
<b>Diagnostic sensitivity</b>	
<b>Proportion of infected/infested samples tested positive compared to results from the standard test, see appendix 2 of PM 7/98</b>	N/A standard test used
<b>Standard test(s)</b>	not relevant
<b>Analytical specificity - inclusivity</b>	
<b>Number of strains/populations of target organisms tested</b>	18 Tomato yellow leaf curl virus - mild (TYLCV) G. Anfoka - origine: Jordanië RefV_TYLCV_01 Tomato yellow leaf curl Sardinia virus (TYLCSV) G. Anfoka - origine: Jordanië RefV_TYLCV_02 Tomato yellow leaf curl virus - mild (TYLCV) DSMZ - PV-0560 - origine Israël RefV_TYLCV_03 Tomato yellow leaf curl Sardinia virus (TYLCSV) DSMZ - PV-0596 - origine Spanje RefV_TYLCV_04 Tomato yellow leaf curl Sardinia virus (TYLCSV) G. Anfoka - origine: Jordanië RefV_TYLCV_05 Tomato yellow leaf curl virus - mild+Israël (TYLCV) G. Anfoka - origine: Jordanië RefV_TYLCV_06 Tomato yellow leaf curl virus (TYLCV) Euphresco THREE - MAF-NZ - non NZ origin RefV_TYLCV_07 Tomato yellow leaf curl virus (TYLCV) Euphresco EIGHT - ARI-CY - Cyprus RefV_TYLCV_08 Tomato yellow leaf curl virus (TYLCV) Euphresco B - FERA UK - Spanje RefV_TYLCV_09 Tomato yellow leaf curl virus (TYLCV) Euphresco FOURTEEN - PPS-NI -CY - Nederland RefV_TYLCV_10 Tomato yellow leaf curl virus (TYLCV) Euphresco E -FERA UK - Spanje RefV_TYLCV_11 Tomato yellow leaf curl virus Israël (TYLCV) Murad Ghanim, Israël RefV_TYLCV_12 Tomato yellow leaf curl virus - mild (TYLCV) DSMZ - PV-0588 RefV_TYLCV_13 Tomato yellow leaf curl virus - mild (TYLCV) DSMZ - PV-0561 RefV_TYLCV_14 Tomato yellow leaf curl virus - mild (TYLCV) BIORÉBA - 223897 RefV_TYLCV_15 Tomato yellow leaf curl virus (TYLCV) Accotto - Italie - 02/02/10 RefV_TYLCV_16 Tomato yellow leaf curl virus (TYLCV) Accotto - Italie - 02/02/10 RefV_TYLCV_17 Tomato yellow leaf curl virus (TYLCV) Verlodt - Tunesie - 02/08/10 RefV_TYLCV_18 Tomato yellow leaf curl virus

	(TYLCV) DSMZ - PV - 0595 RefV_TYLCV_19
<b>Specificity value</b>	
<b>Analytical specificity - exclusivity</b>	
<b>Number of non-target organisms tested</b>	8 Pepino mosaic virus (PepMV) Tomato apical stunt viroid (TASVd) Potato spindle tuber viroid (PSTVd) Tomato spotted wilt virus (TSWV) Cucumber vein yellowing virus (CVYV) Curcubit yellow stunt disorder virus (CYSDV) Tomato infectious chlorosis virus (TiCV) Tomato chlorosis virus (ToCV)
<b>Specificity value</b>	ELISA: possible cross reaction with other begomoviruses qPCR: no cross reaction known
<b>Diagnostic Specificity</b>	
<b>Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test</b>	100%
<b>Reproducibility</b>	
<b>Provide the calculated % of agreement for a given level of the pest (see PM 7/98)</b>	100 % (at low - medium - high concentration)
<b>Repeatability</b>	
<b>Provide the calculated % of agreement for a given level of the pest (see PM 7/98)</b>	100 (at low - medium - high concentration)
<b>Test performance study</b>	
<b>Test performance study?</b>	yes
<b>Brief details of the test performance study and its output. It available, link to published article/report</b>	qPCR: EUPHRESCO project report ""Validation of diagnostic methods for the detection and identification of whitefly transmitted viruses of regulatory or quarantine concern to the EU.""
<b>Other information</b>	
<b>Any other information considered useful</b>	Robustness - also tested are: Influence of sub sampling (different plant parts) Influence of the place in the ELISA plate Buffer and incubation temperature (sample, AB) Dilution of the controls
The following complementary files are available online:	<ul style="list-style-type: none"> <li>• <a href="#">Validation report real-time PCR</a></li> <li>• <a href="#">Validation report TYLCV ELISA</a></li> </ul>

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