

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.

Laboratory contact details	ILVO Institute for Agricultural and Fisheries Research Burg. Van Gansberghelaan 96, 9820 Merelbeke - Melle, Belgium
Short description of the test	DAS-ELISA (screening) and one-step real-time PCR test for Tomato spotted wilt virus
Date, reference of the validation report	2018-02-12 - last version F16_V08; F16_V12
Validation process according to EPPO Standard PM7/98?	yes
Is the lab accredited for this test?	yes
Was the validated data generated in the framework of a project?	no
Description of the test	
Organism(s)	Tomato spotted wilt virus / Orthotospovirus tomatomaculae (TSWV00)
Detection / identification	detection and identification
Matrix(ces) tested	Leaves
Plant species tested	Chrysanthemum sp., Solanum lycopersicum
Method(s)	Molecular Extraction DNA RNA Molecular real time RT PCR Serological DASI-ELISA
Method: Molecular Extraction DNA RNA	
Reference of the test description	
Other information	
Method: Molecular real time RT PCR	
Reference of the test description	
As or adapted from an EPPO diagnostic protocol	yes
EPPO Diagnostic Protocol name	PM 7/034 Tomato spotted wilt, Impatiens necrotic spot and Watermelon silver mottle tospoviruses (version 1)
Name of the test	Taqman real time fluorescent RT-PCR (Boonham et al. 2001)

Is the test modified compared to the reference test	no
Other information	
Reaction type	Duplex - Probe
Other details on the test	qPCR: Boonham et al 2002: The detection of Tomato spotted wilt virus (TSWV) in individual thrips using real time fluorescent RT-PCR (TaqMan). Journal of Virological Methods 101 (2002) 37-48.
Method: Serological DASi-ELISA	
Reference of the test description	
As or adapted from an EPPO diagnostic protocol	yes
EPPO Diagnostic Protocol name	PM 7/034 Tomato spotted wilt, Impatiens necrotic spot and Watermelon silver mottle tospoviruses (version 1)
Name of the test	Triple Antibody Sandwich TAS-ELISA (based on Clark & Adams, 1997)
As or adapted from an IPPC diagnostic protocol	no
Is the test modified compared to the reference test	no
Kit	
Is a kit used	yes
Manufacturer name	DSMZ
Specify the kit used	Tomato spotted wilt virus (TAS-ELISA) TSWV (RT-0105-0106/3)
Kit used following the manufacturer's instructions?	
Other information	
Are the performance characteristics included in the EPPO diagnostic protocol?	no
Performance Criteria :	
Organism 1.:	Orthotospovirus tomatomaculae(TSWV00)
Analytical specificity - inclusivity	
Number of strains/populations of target organisms tested	6 Chrysanthemum Ingelmunster_2011; België RefV_TSWV_01 Chrysanthemum cv. Ludo Sleidinge_2011; België RefV_TSWV_02 Phalaenopsis_2009 Lochristi; België RefV_TSWV_03 Tomato Univ. Plovdiv, Bulgarije 2009 RefV_TSWV_04 Bell pepper Univ. Plovdiv, Bulgarije 2009 RefV_TSWV_05 Tomato spotted wilt virus (TSWV) - DCP 2013, isolate tomato, Belgium RefV_TSWV_06
Specificity value	
Analytical specificity - exclusivity	

Number of non-target organisms tested	12 CSVd chrysanthemum ToMV tomato TRSV tomato CMV tomato CSNV chrysanthemum CVB chrysanthemum INSV Monstera PepMV tomato PVY tomato TBRV potato TMV tobacco TYLCV tomato WSMoV tomato
Specificity value	Cross react with other tospoviruses (eg Chrysanthemum stem necrosis virus)
Cross-reacts with	Chrysanthemum stem necrosis virus
<u>Reproducibility</u>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100%
<u>Repeatability</u>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100%
Test performance study	
Test performance study?	no

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