

**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	Institute for Sustainable Plant Protection via Amendola, 122/D, 70126 Bari, Italy
Short description of the test	Detection of <i>Xylella fastidiosa</i> in perennial host species by ELISA
Date, reference of the validation report	2014-09-01 - 2014-09 and 2015-07 - Maria Saponari, Giuliana Loconsole, Oriana Potere, Donato Boscia, 2014 and 2015. DETECTION OF XYLELLA FASTIDIOSA, INTERLABORATORY VALIDATION - MOLECULAR AND SEROLOGICAL METHODS
Link to other validation data	- 2014-09 and 2015-07 - Maria Saponari, Giuliana Loconsole, Oriana Potere, Donato Boscia, 2014 and 2015. DETECTION OF XYLELLA FASTIDIOSA, INTERLABORATORY VALIDATION - MOLECULAR AND SEROLOGICAL METHODS Detection of <i>Xylella fastidiosa</i> in perennial host species by PCR
Validation process according to EPPO Standard PM7/98?	no
Is the lab accredited for this test?	yes
Was the validated data generated in the framework of a project?	
If yes, please specify	
Description of the test	
Organism(s)	<i>Xylella fastidiosa</i> (XYLEFA)
Detection / identification	detection
Matrix(ces) tested	Leaves leaf petiols
Plant species tested	<i>Acacia saligna</i> , <i>Citrus</i> sp., <i>Nerium oleander</i> , <i>Olea europaea</i> , <i>Polygala myrtifolia</i> , <i>Prunus avium</i> , <i>Prunus dulcis</i> , <i>Quercus ilex</i> , <i>Vitis</i> sp.
Method(s)	Serological DAS-ELISA Serological DAS-ELISA (2)
Method: Serological DAS-ELISA	
Reference of the test description	
As or adapted from an EPPO diagnostic protocol	no

New test being considered for inclusion in the next version of the EPPO diagnostic protocol?	
As or adapted from an IPPC diagnostic protocol	
Is the test modified compared to the reference test	
Kit	
Is a kit used	yes
Manufacturer name	LOEWE
Specify the kit used	Xylella fastidiosa grapevine isolate DAS ELISA Cat. No. 07119
Kit used following the manufacturer's instructions?	
Other information	
Reaction type	
Other details on the test	Loconsole, G., Potere, O., Boscia, D., Altamura, G., Djelouah, K., Elbeaino, T., Frasheri, D., Lorusso, D., Palmisano, F., Pollastro, P., Silletti, M. R., Trisciuzzi, N., Valentini, F., Savino V. & Saponari, M. (2014a). Detection of Xylella fastidiosa in olive trees by serological and molecular methods. Journal of Plant Pathology, 96, 7-14; Loconsole, G., Potere, O., Elbeaino, T., Frasheri, D., Frisullo, S., Palmisano, P., Boscia, D. & Saponari, M. (2014b). Interlaboratory validation of molecular and serological diagnosis of Xylella fastidiosa strain CoDiRO in susceptible host plants. International Symposium on the European Outbreak of Xylella fastidiosa in Olive, Journal of Plant Pathology, 96, S4.100.
Method: Serological DAS-ELISA (2)	
Reference of the test description	
As or adapted from an EPPO diagnostic protocol	
New test being considered for inclusion in the next version of the EPPO diagnostic protocol?	
As or adapted from an IPPC diagnostic protocol	
Is the test modified compared to the reference test	
Kit	
Is a kit used	yes
Manufacturer name	AGRITEST
Specify the kit used	DAS ELISA KIT Xylella fastidiosa
Kit used following the manufacturer's instructions?	
Other information	
Reaction type	

Other details on the test	Loconsole, G., Potere, O., Boscia, D., Altamura, G., Djelouah, K., Elbeaino, T., Frasheri, D., Lorusso, D., Palmisano, F., Pollastro, P., Silletti, M. R., Trisciuzzi, N., Valentini, F., Savino V. & Saponari, M. (2014a). Detection of Xylella fastidiosa in olive trees by serological and molecular methods. Journal of Plant Pathology, 96, 7-14; Loconsole, G., Potere, O., Elbeaino, T., Frasheri, D., Frisullo, S., Palmisano, P., Boscia, D. & Saponari, M. (2014b). Interlaboratory validation of molecular and serological diagnosis of Xylella fastidiosa strain CoDiRO in susceptible host plants. International Symposium on the European Outbreak of Xylella fastidiosa in Olive, Journal of Plant Pathology, 96, S4.100.
Are the performance characteristics included in the EPPO diagnostic protocol?	yes
Performance Criteria :	
Organism 1.:	Xylella fastidiosa(XYLEFA)
<u>Analytical sensitivity</u>	
What is smallest amount of target that can be detected reliably?	up to 10 ⁴ cfu/ml with the kits Agritest and Loewe, using dilutions ranging from 10 ⁷ to 10 CFU/ml, prepared by spiking the inactivated bacterial culture in olive ELISA homogenate
<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	100%
Standard test(s)	76 obtained positive samples/76 expected positive samples
<u>Analytical specificity - inclusivity</u>	
Number of strains/populations of target organisms tested	
Specificity value	
<u>Analytical specificity - exclusivity</u>	
Number of non-target organisms tested	
Specificity value	
Cross reacts with	
<u>Diagnostic Specificity</u>	
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	100%
Specify the test(s)	84 obtained negative samples/84 expected negative samples
<u>Reproducibility</u>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100%

Repeatability	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100%
Test performance study	
Test performance study?	no
Brief details of the test performance study and its output. It available, link to published article/report	
Other information	
Any other information considered useful	Validation of the ELISA protocol was carried out by the Laboratories listed below, under the supervision of the reference laboratory IPSP-CNR e DiSSPA-UNIBA: • IPSP-CNR: Istituto per la Protezione Sostenibile delle Piante CNR, UOS Bari (ITALY); • DiSSPA-UNIBA: Dipartimento di Scienze del Suolo, della Pianta e degli Alimenti, Università degli Studi Aldo Moro, Bari (Italy); • CRSFA: Centro di Ricerca, Sperimentazione e Formazione in Agricoltura Basile Caramia, Locorotondo (BA), Italy; • IAMB: Istituto Agronomico Mediterraneo, Valenzano (BA), Italy; • Dipartimento di Scienze Agroambientali, Chimica e Difesa Vegetale - Università degli Studi di Foggia, (ITALY). A panel of blind samples was distributed.
The following complementary files are available online:	<ul style="list-style-type: none"> • protocols for diagnosis of Xylella fastidiosa • Report interlaboratory validation 2015

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