

**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	Naktuinbouw Sotaweg 22, 2371 GD Roelofarendsveen, Netherlands
Short description of the test	Isolation of <i>Xanthomonas</i> sp. from tomato and pepper seeds
Date, reference of the validation report	2012-01-26 - Validation report for the isolation of <i>Xanthomonas</i> sp. from tomato and pepper seeds, Naktuinbouw, 2012
Validation process according to EPPO Standard PM7/98?	yes
Is the lab accredited for this test?	no
Was the validated data generated in the framework of a project?	
Description of the test	
Organism(s)	<i>Xanthomonas vesicatoria</i> (XANTVE) <i>Xanthomonas euvesicatoria</i> pv. <i>euvesicatoria</i> (XANTEU) <i>Xanthomonas euvesicatoria</i> pv. <i>perforans</i> (XANTPF) <i>Xanthomonas hortorum</i> pv. <i>gardneri</i> (XANTGA)
Detection / identification	detection
Method(s)	Isolation Isolation (2)
Method: Isolation	
Reference of the test description	
As or adapted from an EPPO diagnostic protocol	yes
New test being considered for inclusion in the next version of the EPPO diagnostic protocol?	no
EPPO Diagnostic Protocol name	PM 7/110 <i>Xanthomonas</i> spp. (<i>Xanthomonas euvesicatoria</i> , <i>Xanthomonas gardneri</i> , <i>Xanthomonas perforans</i> , <i>Xanthomonas vesicatoria</i>) causing bacterial spot of tomato and sweet pepper (version 1)
Name of the test	Isolation from seed on mTMB
As or adapted from an IPPC diagnostic protocol	no

Is the test modified compared to the reference test	yes This is a modification of the current EPPO DP, NSCCA medium was used.
Other information	
Other details on the test	McGuire, R.G., Jones, J.B., Sasser, M. (1986). Tween media for Semiselective Isolation of <i>Xanthomonas campestris</i> pv. <i>vesicatoria</i> from soil and plant material. <i>Plant Disease</i> 70, 887-891. S
Method: Isolation (2)	
Reference of the test description	
As or adapted from an EPPO diagnostic protocol	yes
New test being considered for inclusion in the next version of the EPPO diagnostic protocol?	no
EPPO Diagnostic Protocol name	PM 7/110 <i>Xanthomonas</i> spp. (<i>Xanthomonas euvesicatoria</i> , <i>Xanthomonas gardneri</i> , <i>Xanthomonas perforans</i> , <i>Xanthomonas vesicatoria</i>) causing bacterial spot of tomato and sweet pepper (version 1)
Name of the test	Isolation from seed on mMXV
As or adapted from an IPPC diagnostic protocol	no
Is the test modified compared to the reference test	yes This is a modification of the current EPPO DP, NSCCA medium was used.
Other information	
Other details on the test	Sijam, K., Chang, C.J., Gitaitis, R.D. (1991). An agar medium for the isolation and identification of <i>Xanthomonas campestris</i> pv. <i>vesicatoria</i> from seed. <i>Phytopathology</i> 81, 831-834.
Performance Criteria :	
Organism 1.:	<i>Xanthomonas vesicatoria</i>(XANTVE)
Analytical sensitivity	
What is smallest amount of target that can be detected reliably?	24 CFU/ml
Analytical specificity - inclusivity	
Number of strains/populations of target organisms tested	14 <i>X. euvesicatoria</i> , two <i>X. gardneri</i> , six <i>X. perforans</i> and six <i>X. vesicatoria</i> isolates were selected for determination of analytical specificity of the dilution plating on the semi-selective media mMXV and mTMB.
Specificity value	Analytical specificity was good. The method was able to detect all tested isolates of the XCV species complex.
Analytical specificity - exclusivity	
Number of non-target organisms tested	three isolates of <i>Clavibacter michiganensis</i> subsp. <i>michiganensis</i> and one isolate of <i>Pseudomonas syringae</i> pv. <i>tomato</i>

Specificity value	no cross reaction The other tested seed borne pathogens <i>Clavibacter michiganensis</i> subsp. <i>michiganensis</i> and <i>Pseudomonas syringae</i> pv. <i>tomato</i> were not able to grow on the semi-selective media mMXV and mTMB.
Reproducibility	
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%
Organism 2.:	<i>Xanthomonas euvesicatoria</i> pv. <i>euvesicatoria</i>(XANTEU)
Analytical sensitivity	
What is smallest amount of target that can be detected reliably?	53 CFU/ml
Analytical specificity - inclusivity	
Number of strains/populations of target organisms tested	14 <i>X. euvesicatoria</i> , two <i>X. gardneri</i> , six <i>X. perforans</i> and six <i>X. vesicatoria</i> isolates were selected for determination of analytical specificity of the dilution plating on the semi-selective media mMXV and mTMB.
Specificity value	Analytical specificity was good. The method was able to detect all tested isolates of the XCV species complex.
Analytical specificity - exclusivity	
Number of non-target organisms tested	three isolates of <i>Clavibacter michiganensis</i> subsp. <i>michiganensis</i> and one isolate of <i>Pseudomonas syringae</i> pv. <i>tomato</i>
Specificity value	no cross reaction The other tested seed borne pathogens <i>Clavibacter michiganensis</i> subsp. <i>michiganensis</i> and <i>Pseudomonas syringae</i> pv. <i>tomato</i> were not able to grow on the semi-selective media mMXV and mTMB.
Reproducibility	
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%
Organism 3.:	<i>Xanthomonas euvesicatoria</i> pv. <i>perforans</i>(XANTPF)
Analytical sensitivity	
What is smallest amount of target that can be detected reliably?	334 CFU/ml
Analytical specificity - inclusivity	

Number of strains/populations of target organisms tested	14 X. euvesicatoria, two X. gardneri, six X. perforans and six X. vesicatoria isolates were selected for determination of analytical specificity of the dilution plating on the semi-selective media mMXV and mTMB.
Specificity value	Analytical specificity was good. The method was able to detect all tested isolates of the XCV species complex.
<u>Analytical specificity - exclusivity</u>	
Number of non-target organisms tested	three isolates of Clavibacter michiganensis subsp. michiganensis and one isolate of Pseudomonas syringae pv. tomato
Specificity value	The other tested seed borne pathogens Clavibacter michiganensis subsp. michiganensis and Pseudomonas syringae pv. tomato were not able to grow on the semi-selective media mMXV and mTMB. no cross reaction
<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%
Organism 4.:	Xanthomonas hortorum pv. gardneri(XANTGA)
<u>Analytical sensitivity</u>	
What is smallest amount of target that can be detected reliably?	94 CFU/ml
<u>Analytical specificity - inclusivity</u>	
Number of strains/populations of target organisms tested	14 X. euvesicatoria, two X. gardneri, six X. perforans and six X. vesicatoria isolates were selected for determination of analytical specificity of the dilution plating on the semi-selective media mMXV and mTMB.
Specificity value	Analytical specificity was good. The method was able to detect all tested isolates of the XCV species complex.
<u>Analytical specificity - exclusivity</u>	
Number of non-target organisms tested	three isolates of Clavibacter michiganensis subsp. michiganensis and one isolate of Pseudomonas syringae pv. tomato
Specificity value	no cross reaction The other tested seed borne pathogens Clavibacter michiganensis subsp. michiganensis and Pseudomonas syringae pv. tomato were not able to grow on the semi-selective media mMXV and mTMB.
<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
The following complementary files are available online:	
	<ul style="list-style-type: none"> • Isolation of Xanthomonas sp. from tomato and papeer seeds

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