

**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>	EUPHRESCO PANTOEA STEWARTII , , EUPHRESCO
<b>Short description of the test</b>	Detection of 'Candidatus Phytoplasma mali', 'Ca. P. pyri' and 'Ca. P. prunorum' by real-time PCR
<b>Date, reference of the validation report</b>	2011-05-31 - Not specified
<b>Validation process according to EPPO Standard PM7/98?</b>	yes
<b>Is the lab accredited for this test?</b>	no
<b>Was the validated data generated in the framework of a project?</b>	Euphresco
<b>If yes, please specify</b>	EUPH-PANTOEA
<b>Description of the test</b>	
<b>Organism(s)</b>	Pantoea stewartii subsp. stewartii(ERWIST)
<b>Detection / identification</b>	detection and identification
<b>Method(s)</b>	Serological IF Serological IF (2)
<b>Method: Serological IF</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 7/060 Pantoea stewartii (version 2)
<b>Name of the test</b>	Cell staining according to PM7/97
<b>Is the test modified compared to the reference test</b>	no
<b>Kit</b>	
<b>Is a kit used</b>	yes
<b>Manufacturer name</b>	
<b>Specify the kit used</b>	
<b>Kit used following the manufacturer's instructions?</b>	
<b>Other information</b>	
<b>Other details on the test</b>	Commercial Loewe polyclonal antibodies - IF on seed macerates

<b>Method: Serological IF (2)</b>	
<b>Reference of the test description</b>	
<b>As or adapted from an EPPD diagnostic protocol</b>	yes
<b>EPPD Diagnostic Protocol name</b>	PM 7/060 Pantoea stewartii (version 2)
<b>Name of the test</b>	Cell staining according to PM7/97
<b>Kit</b>	
<b>Is a kit used</b>	yes
<b>Manufacturer name</b>	LINARIS
<b>Specify the kit used</b>	Pantoea stewartii:AP (Pstew), Monoclonal, Conjugated Antibody
Kit used following the manufacturer's instructions?	
<b>Other information</b>	
<b>Other details on the test</b>	Linaris monoclonal antibodies
<b>Are the performance characteristics included in the EPPD diagnostic protocol?</b>	<b>yes</b>
<b>Performance Criteria :</b>	
<b>Organism 1.:</b>	<b>Pantoea stewartii subsp. stewartii(ERWIST)</b>
<b>Analytical sensitivity</b>	
<b>What is smallest amount of target that can be detected reliably?</b>	Linaris: $10^3$ to $10^4$ CFU/mL ; Loewe: $10^3$ CFU/mL on pure culture
<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>	Not tested
<b>Analytical specificity - inclusivity</b>	
<b>Number of strains/populations of target organisms tested</b>	Inclusivity tested with 15 target strains: 100% (bacterial suspension concentrations about $10^7$ bact./mL) Pantoea stewartii subsp. stewartii: CFBP 3167/ NCPPB 2295/ICMP 257ATCC 8199; CFBP1719/ ICPB SS104; CFBP 2502 / NCPPB 449; CFBP 3157/ NCPPB 1553; CFBP 3166 / ICMP 5930; CFBP 3393/ LMG 2716/ PDDCC 270; CFBP 3394/ LMG 2717/ PDDCC 722; CFBP 3395/ LMG 2718/ ATCC 8200; CFBP 3396/ LMG 2719/ PDDCC 5929; CFBP 3445/ NCPPB 3379; CFBP 3517; CFBP 3168; CFBP 3165; CFBP 3169; NCPPB 3253;
<b>Specificity value</b>	Inclusivity + exclusivity Linaris = 100% on 30 target and no-target strains Loewe= 98,6% on 30 target and no-target strains
<b>Analytical specificity - exclusivity</b>	
<b>Number of non-target organisms tested</b>	Exclusivity tested with 15 non-target strains (bacterial suspension concentration of about $10^7$ bact./mL): Linaris: 100% Loewe: 96.2% Clavibacter michiganensis subsp. michiganensis CFBP 4999 /

	LNPV 30.31; Clavibacter michiganensis subsp. nebraskensis CFBP 2405 / LNPV 10.17; Curtobacterium flacumfasciens pv flacumfasciens CFBP 3456 /LNPV 10.24; Erwinia chrisanthemi pv.zea CFBP 2052 Erwinia amylovora CFBP 1232/ NCPPB 683/ ATCC 15580/ CCM 114; Erwinia carotovora subsp. carotovora CFBP 2046; Erwinia carotovora subsp. atroseptica CFBP 1526; Pantoea agglomerans CFBP 3845/ ATCC 27155/ CIP 5751; Pantoea ananas pv. uredovora CFBP 3171 Pseudomonas syringae pv. syringae CFBP 1392; Pseudomonas viridiflava CFBP 1141 / LNPV 3.40; Xanthomonas campestris pv. campestris CFBP 5251 /NCPBP 528; Pantoea stewartii subsp.indologenes CFBP 3614/ ICMP 77 / LMG 2632 / NCPPB 2280; Pseudomonas syringae pv lapsa CFBP 1731; Pseudomonas corrugata CFBP 2431
<b>Specificity value</b>	
<b>Cross reacts with</b>	Erwinia amylovora Pseudomonas corrugata
<b>Repeatability</b>	
<b>Provide the calculated % of agreement for a given level of the pest (see PM 7/98)</b>	100%
<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>	Based on the test performance study with 5 laboratories involved, in the framework of EUPH05 Pantoea stewartii ssp. stewartii, EUPHRESCO Final Report 2010-2011 Linaris: Analytical sensitivity: 95% of agreement at <math><7 \times 10^3 \text{ CFU/mL}</math> Diagnostic sensitivity (10 samples/2repeats/soak/lab): 75 % Diagnostic specificity (5 samples/2repeats/soak/lab): 33 % (cross reaction with P st indologenes) Qualitative Repeatability: 97 % Accuracy: 61% Qualitative Reproducibility (common Linaris and Loewe): 65 % Loewe: Analytical sensitivity: 70% of agreement at <math><7 \times 10^3 \text{ CFU/mL}</math> Diagnostic sensitivity (10 samples/2repeats/soak/lab): 92,5 % Diagnostic specificity (5 samples/2repeats/soak/lab): 95 % (cross reaction with P st indologenes) Qualitative Repeatability: 79 % Accuracy: 93%

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