

EUROPEAN AND MEDITERRANEAN PLANT PROTECTION ORGANIZATION
ORGANISATION EUROPEENNE ET MEDITERRANEENNE POUR LA PROTECTION DES PLANTES
(11-17239)

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.

Target Organism	Plum pox virus Prunus necrotic ringspot virus Prune dwarf virus	
Short description	detection of PPV, PNRV and PDV by DAS-ELISA	
Laboratory contact details	Bavarian State Research Center for Agriculture, Institute for Plant Protection - Phytopathology and Diagnosis Lange Point 10, 85354 Freising, Germany	
Date and reference of the validation report	31/03/2014 - FB 20.02.05/5	
Validation process according to EPPO Standard PM 7/98:	Yes	
Reference of the test description	N/R AA 20.02.02.06 ELISA tests of seeds (stones), leaves, buttons, bark of different Prunus species for PDV, PNRSV and PPV AA 20.02.02.01 Performance of the DAS ELISA for virus detection on plant material	
Is the test the same as described in the EPPO DP?		
Is the lab accredited for this test?	Yes	
Plant species tested (if relevant)	Not relevant	
Matrices tested (if relevant)	Leaf material	
List of methods used		
Method for extraction / isolation / baiting of target organism from matrix		
Molecular methods, e.g. hybridization, PCR and real time PCR		
Serological methods: IF, ELISA, Direct Tissue Blot Immuno Assay	X	DAS-ELISA: Nunc maxisorp plates; antisera Loewe Biochemica, coating 100 µl: PDV Loewe 07051 PPV Loewe 07186 universal PNRSV Loewe 07052
Plating methods: selective isolation		
Bioassay methods: selective enrichment in host plants, baiting, plant test and grafting.		
Pathogenicity test		

Fingerprint methods: protein profiling, fatty acid profiling & DNA profiling		
Morphological and morphometrical methods intended for identification		
Biochemical methods: e.g. enzyme electrophoresis, protein profiling		
Other		
<u>Analytical sensitivity (= limit of detection)</u>		
What is smallest amount of target that can be detected reliably?	For PPV and PDV: 100% correct positive up to a dilution of 1:40960 (highest dilution tested; dilution in negative extract from tobacco). Analytical sensitivity is lower for PNRSV (100% correct positive up to 1:2560, dilution in negative extract from tobacco)	
<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	Not evaluated	
Specify the standard test		
<u>Analytical specificity</u>		
Specificity value	Not evaluated	
Number of strains/populations of target organisms tested	1 positive PDV control Loewe Biochemica 1 positive PNRSV control PV-PC-0962 DSMZ 1 positive PPV control PV-PC-0305 DSMZ	
Number of non-target organisms tested		
Cross reacts with (specify the species)		
<u>Diagnostic Specificity</u>		
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	Not evaluated	
Specify the standard test		
<u>Reproducibility</u>		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	Two technical assistants: (dilution in extract from negative tobacco) 100% for PDV and PPV up to and including dilution 1:40960 (highest dilution tested,) 100 % for PNRSV up to and including diluton 1:2560	
<u>Repeatability</u>		
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	100% for PDV and PPV up to diluton 1:40960 (highest dilution tested, dilution in negative extract from tobacco). 100 % for PNRSV up o diluton 1:2560 (dilution in negative extract from tobacco))	

Test performance study	
Test performance study?	No
Include brief details of the test performance study and its output. If available, provide a link to published article/report	
Other information	
Any other information considered useful e.g. robustness, ease of performing the test, etc.	
The following complementary files are available online:	<ul style="list-style-type: none"> • FB 20.02.05_5 Ergänzung Methodenvvalidierung_PDV PNRSV PPV