

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	EUPH DNA Barcoding , , EUPHRESKO
Short description of the test	DNA Barcoding - Optimizing and validating DNA barcoding protocols for Nematode
Date, reference of the validation report	2016-06-30 - Final_report_DNA barcoding.doc
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?	Euphresco
If yes, please specify	EUPHRESKO DNA Barcoding - Optimizing and validating DNA barcoding protocols for plant pests
Description of the test	
Organism(s)	Aphelenchoides fragariae(APLOFR) Meloïdogyne chitwoodi(MELGCH) Ditylenchus dipsaci(DITYDI) Bursaphelenchus xylophilus(BURSXY) Aphelenchoïdes besseyi(APLOBE)
Detection / identification	identification
Matrix(ces) tested	Specimen
Plant species tested	
Method(s)	Molecular other
Method: Molecular other	
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?	
EPPO Diagnostic Protocol name	PM 7/129 DNA barcoding as an identification tool for a number of regulated pests (version 1)
Name of the test	
As or adapted from an IPPC diagnostic protocol	no

Is the test modified compared to the reference test	no
Kit	
Is a kit used	no
Other information	
Reaction type	
Other details on the test	Phusion® High-Fidelity (New England Biolabs) See report for details about the primers & protocols
Are the performance characteristics included in the EPPO diagnostic protocol?	yes
Performance Criteria :	
Organism 1.:	Aphelenchoides fragariae(APLOFR)
Analytical sensitivity	
What is smallest amount of target that can be detected reliably?	See report
Diagnostic sensitivity	
Proportion of infected/infested samples tested positive compared to results from the standard test, see appendix 2 of PM 7/98	89% (100% after re-analysing the consensus sequence data provided by TPS participants)
Standard test(s)	
Analytical specificity - inclusivity	
Number of strains/populations of target organisms tested	See report
Specificity value	
Analytical specificity - exclusivity	
Number of non-target organisms tested	See report
Specificity value	
Cross reacts with	
Diagnostic Specificity	
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	
Specify the test(s)	
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)	
Organism 2.:	Meloidogyne chitwoodi(MELGCH)
Analytical sensitivity	

What is smallest amount of target that can be detected reliably?	See report
<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)	
<u>Analytical specificity - inclusivity</u>	
Number of strains/populations of target organisms tested	See report
Specificity value	
<u>Analytical specificity - exclusivity</u>	
Number of non-target organisms tested	See report
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	
Specify the test(s)	
<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)	
Organism 3.:	Ditylenchus dipsaci(DITYDI)
<u>Analytical sensitivity</u>	
What is smallest amount of target that can be detected reliably?	See report
<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)	
<u>Analytical specificity - inclusivity</u>	
Number of strains/populations of target organisms tested	See report
Specificity value	
<u>Analytical specificity - exclusivity</u>	
Number of non-target organisms tested	See report

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	
Specify the test(s)	
<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)	
Organism 4.:	Bursaphelenchus xylophilus(BURSXY)
<u>Analytical sensitivity</u>	
What is smallest amount of target that can be detected reliably?	See report
<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)	
<u>Analytical specificity - inclusivity</u>	
Number of strains/populations of target organisms tested	See report
Specificity value	
<u>Analytical specificity - exclusivity</u>	
Number of non-target organisms tested	See report
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	
Specify the test(s)	
<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)	

Organism 5.:	Aphelechooides besseyi(APLOBE)
Analytical sensitivity	
What is smallest amount of target that can be detected reliably?	See report
Diagnostic sensitivity	
Proportion of infected/infested samples tested positive compared to results from the standard test, see appendix 2 of PM 7/98	89% (100% after re-analysing the consensus sequence data provided by TPS participants)
Standard test(s)	
Analytical specificity - inclusivity	
Number of strains/populations of target organisms tested	See report
Specificity value	
Analytical specificity - exclusivity	
Number of non-target organisms tested	See report
Specificity value	
Cross reacts with	
Diagnostic Specificity	
Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test	
Specify the test(s)	
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)	
Test performance study	
Test performance study?	yes
Brief details of the test performance study and its output.It available, link to published article/report	Test performance study involving 11 laboratories. See report for more information.
Other information	
Any other information considered useful	
The following complementary files are available online:	<ul style="list-style-type: none"> • EUPHRESCO DNA Barcoding - report

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