

**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>	Anses Plant Health Laboratory - Nematology Unit Domaine de la Motte au Viconte BP 35327, 35653 Le Rheu, France
<b>Short description of the test</b>	identification of <i>Meloidogyne graminicola</i> by Molecular real time PCR in juveniles
<b>Date, reference of the validation report</b>	2024-08-21 - Identification of <i>Meloidogyne graminicola</i> by real-time PCR Mattos et al., 2019 on isolated juveniles
<b>Link to other validation data</b>	- Identification of <i>Meloidogyne graminicola</i> by real-time PCR Htay et al 2016 on isolated juveniles identification of <i>Meloidogyne graminicola</i> by Molecular real time PCR in juveniles
<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>	EURL
<b>If yes, please specify</b>	EU funded project EURLs-EURCs 2023-2024 (grant Project 101143591)
<b>Description of the test</b>	
<b>Organism(s)</b>	<i>Meloidogyne graminicola</i> (MELGGC)
<b>Detection / identification</b>	identification
<b>Matrix(ces) tested</b>	Specimen Juveniles
<b>Method(s)</b>	Molecular real time PCR
<b>Method: Molecular real time PCR</b>	
<b>Reference of the test description</b>	
<b>As or adapted from an EPPO diagnostic protocol</b>	no
<b>New test being considered for inclusion in the next version of the EPPO diagnostic protocol?</b>	yes
<b>As or adapted from an IPPC diagnostic protocol</b>	no
<b>Reference of the test</b>	Mattos et al., 2019
<b>Is the test modified compared to the</b>	yes Adapted for a real-time PCR

reference test	
<b>Kit</b>	
Is a kit used	no
<b>Other information</b>	
Reaction type	Simplex
Other details on the test	-The test was developed by Mattos et al., 2019, and further adapted by INIAV during an EURL TPS (Report 22MG), and validated by the EURL for Plant Parasitic Nematode
<b>Performance Criteria :</b>	
Organism 1.:	<b>Meloidogyne graminicola(MELGGC)</b>
<b>Analytical sensitivity</b>	
What is the smallest amount of target that can be detected reliably?	1 nematode (J2) 100%
<b>Analytical specificity - inclusivity</b>	
Number of strains/populations of target organisms tested	one population of M. oryzae from Brazil
Specificity value	100%
<b>Analytical specificity - exclusivity</b>	
Number of non-target organisms tested	2 populations (Italy and Philippines) of M. graminicola. However, in conventional PCR was evaluated on M. naasi, M. incognita, M. javanica, and M. salasi.
Specificity value	100%, no amplification or a Ct value >35 was obtained for the tested populations.
<b>Reproducibility</b>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	8 replicates were analyzed in 2 different trials, performed on different days and/or using 2 different real-time PCR thermocyclers: 100% for 1, 2, 5 and 10 juveniles (J2) of M. oryzae (8 replicates x 2 PCR trials x 4 modalities = 64 tests).
<b>Repeatability</b>	
Provide the calculated % of agreement for a given level of the pest (see PM 7/98)	8 replicates: 100% for 1, 2, 5 and 10 juveniles (J2) of M. oryzae were analyzed in 3 different tested trials (8 replicates x 3 PCR trials x 4 modalities = 96 tests)
<b>Test performance study</b>	
Test performance study?	yes
Brief details of the test performance study and its output. It available, link to published article/report	TEST PERFORMANCE STUDY REPORT 22MG Identification of Meloidogyne graminicola by molecular conventional PCR Mattos et al 2019 (oryzae primers) in juveniles
<b>Other information</b>	
Any other information considered useful	Report available on the EURL website for the NRLs or available on request to the EURL.

