

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.

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| Laboratory contact details | National Institute of Biology, Department of Biotechnology and Systems Biology Vecna pot 121, 1000 Ljubljana, Slovenia |
| Short description of the test | Detection of <i>Ralstonia solanacearum</i> by egl LAMP in plant material |
| Date, reference of the validation report | 2017-02-09 - Dreo, T., 2017. Summary of validation data on egl LAMP for <i>Ralstonia solanacearum</i> (No. D0004/17), Report on Suitability Testing. National Institute of Biology, Ljubljana. |
| 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 | |
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| Organism(s) | <i>Ralstonia solanacearum</i> species complex (RALSSO) |
| Detection / identification | detection |
| Method(s) | Molecular LAMP |
| Method: Molecular LAMP | |
| Reference of the test description | |
| Other information | |
| Other details on the test | LAMP egl |
| Are the performance characteristics included in the EPPO diagnostic protocol? | no |
| Performance Criteria : | |
| Organism 1.: | <i>Ralstonia solanacearum</i> species complex(RALSSO) |
| Analytical sensitivity | |
| What is smallest amount of target that can be detected reliably? | 10E4 cells/mL (25 cells per LAMP reaction) when tested on strains belonging to phylotypes I and III, and a sensitivity limit of 10E5–10E6 cells/mL for strains from phylotypes IIA, IIB and IV; 10E5 cells/mL in potato tubers (cores of 200) as tested on three standard curves. |

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| Diagnostic sensitivity | |
| 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) | Potato cores. The official testing scheme (EC 98/57) including screening tests, isolation on media, identification tests and pathogenicity testing with re-isolation and identification for positive samples. |
| Analytical specificity - inclusivity | |
| Number of strains/populations of target organisms tested | 88 strains of RSSC |
| Specificity value | 99 % (1 false negative, no false positives) |
| Analytical specificity - exclusivity | |
| Number of non-target organisms tested | 26 |
| Specificity value | no cross-reactions observed |
| Diagnostic Specificity | |
| Proportion of uninfected/uninfested samples (true negatives) testing negative compared to results from a standard test | 100% |
| Specify the test(s) | Immunofluorescence and real-time PCR (Weller et al., 2000) for negative samples. |
| Reproducibility | |
| Provide the calculated % of agreement for a given level of the pest (see PM 7/98) | 100 % detection for samples with at least 10E4 copies of Rs DNA or more. |
| Repeatability | |
| Provide the calculated % of agreement for a given level of the pest (see PM 7/98) | 100 % using different machines (SmartCycler, Roche Light Cycler, Genie II). |
| Test performance study | |
| Test performance study? | no |
| Other information | |
| Any other information considered useful | The test is proposed as identification test for pure cultures. While the validation data indicates that it may well detect concentrations of <i>R. solanacearum</i> usually seen in latently infected samples there is not sufficient data on the R.s. concentrations encountered in routine testing. |
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| The following complementary files are available online: | <ul style="list-style-type: none"> • Summary of Rs LAMP validation data |

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