

Validation report

qPCR Rs set

Article No.: 869600 (qPCR Rs set 96) / 869200 (qPCR Rs set 192)

General information:

Target Pathogen	RSSC (<i>Ralstonia solanacearum</i> species complex)
Genus	<i>Ralstonia</i>
Method	Real-Time qPCR, TaqMan (Multiplex)

Technical information:

Fluorophores	HEX: Rs ROX: 18S rRNA (IC)
Cycling program	➤ 95°C for 5 min (Hot Start) ➤ 45 Cycles: ○ 95°C for 10 sec (Denaturation) ○ 60°C for 40 sec (Annealing / Extension)
Controls	Internal positive control (IPC): Host 18S rRNA gene sequence Negative control (NC): plant DNA Positive control (PC): Rs-infected plant extracts

Host matrix:

Tested plant material	Leaf, tuber
Tested species infected	<i>Musa</i> sp. (Banana) <i>Rosa</i> sp. (Rose) <i>Solanum lycopersicum</i> (Tomato) <i>Solanum tuberosum</i> (Potato) <i>Zingiber officinale</i> (Ginger)

Specificity:

Analytical Specificity	100%
Number of tested samples from target organisms (True Positives)	16
Diagnostic Specificity	100%
Number of tested samples non-target organism (True Negatives)	>100
Detected isolates / geographic regions (Inclusivity)	
R. pseudosolanacearum (Phylotyp I, Switzerland) R. pseudosolanacearum (Phylotype I, Curcuma, Peru) R. pseudosolanacearum (Phylotype I, Rose, Netherlands) R. pseudosolanacearum (Phylotype I, Rose, Netherlands) R. pseudosolanacearum (Phylotype I, GBBC 581) R. pseudosolanacearum (Phylotype I, CFBP_707, Tomato, Tahiti) R. pseudosolanacearum (Phylotype I, CFBP_8427, Tomato, Indonesia) R. pseudosolanacearum (Phylotype I, GBBC_1112, Potato, Belgium) R. pseudosolanacearum (Phylotype I, GBBC_1172, Potato, China) R. pseudosolanacearum (Phylotype I, GBBC_1200, Potato, Mali) R. pseudosolanacearum (Phylotype I, GBBC_1202, Potato, Mali) R. pseudosolanacearum (Phylotype I, GBBC_1331, Ginger, China) R. pseudosolanacearum (Phylotype I, GBBC_1334, Eggplant, China)	

R. pseudosolanacearum (Phylotype I, GBBC_1338, Eggplant, China)
 R. pseudosolanacearum (Phylotype I, GBBC_1679, Potato, Congo)
 R. pseudosolanacearum (Phylotype I, GBBC_1681, Potato, Zambia)
 R. pseudosolanacearum (Phylotype I, GBBC_3080, Rose, Belgium)
 R. pseudosolanacearum (Phylotype I, LMG_2297, Coastal She-oak, Mauritius)
 R. pseudosolanacearum (Phylotype I, LMG_26750, Potato, Nepal)
 R. pseudosolanacearum (Phylotype I, LMG_5839, Ginger, USA)
 R. pseudosolanacearum (Phylotype I, NCPPB_4000, Ginger, Thailand)
 R. pseudosolanacearum (Phylotype I, CFBP_1960, Bell Pepper, Algeria)
 R. pseudosolanacearum (Phylotype I, CFBP_6442, Mulberry, China)
 R. pseudosolanacearum (Phylotype I, CFBP_8426, Tomato, Taiwan)
 R. pseudosolanacearum (Phylotype I, NCPPB_1484, Crane Flower, Mauritius)
 R. pseudosolanacearum (Phylotype I, NCPPB_3190, Tomato, Malaysia)
 R. pseudosolanacearum (Phylotype I, NCPPB_3793, Potato, Java)
 R. solanacearum (Phylotyp II, Switzerland)
 R. solanacearum (Phylotype II, Potato, Switzerland)
 R. solanacearum (Phylotype II, Potato, Germany)
 R. solanacearum (Phylotype II, Potato, Germany)
 R. solanacearum (Phylotype II, Potato, Germany)
 R. solanacearum (Phylotype II, GBBC 504)
 R. solanacearum (Phylotype II, GBBC 507)
 R. solanacearum (Phylotype II, GBBC 583)
 R. solanacearum (Phylotype II, GBBC 588)
 R. solanacearum (Phylotype II, GBBC 594)
 R. solanacearum (Phylotype II, GBBC 603)
 R. solanacearum (Phylotype II, GBBC 608)
 R. solanacearum (Phylotype II, GBBC 665)
 R. solanacearum (Phylotype II, GBBC 666)
 R. solanacearum (Phylotype II, GBBC 3335)
 R. solanacearum (Phylotype II, GBBC 3360)
 R. solanacearum (Phylotype II, GBBC 3362)
 R. solanacearum (Phylotype II, GBBC 3519)
 R. solanacearum (Phylotype II, GBBC_1240)
 R. solanacearum (Phylotype II, GBBC_1685)
 R. solanacearum (Phylotype II, GBBC_765)
 R. pseudosolanacearum (Phylotyp III, NCPPB 3181, Gambia)
 R. pseudosolanacearum (Phylotype III, CFBP_4961, Geranium, Réunion)
 R. pseudosolanacearum (Phylotype III, CFBP_7062, Potato, Cameroon)
 R. pseudosolanacearum (Phylotype III, GBBC_1122, Potato, Guinea)
 R. pseudosolanacearum (Phylotype III, GBBC_1129, Potato, Guinea)
 R. pseudosolanacearum (Phylotype III, GBBC_1210, Potato, Guinea)
 R. pseudosolanacearum (Phylotype III, GBBC_1222, Potato, Guinea)
 R. pseudosolanacearum (Phylotype III, GBBC_1238, Guinea)
 R. pseudosolanacearum (Phylotype III, GBBC_1259, Eggplant, Guinea)
 R. pseudosolanacearum (Phylotype III, GBBC_1950, Burundi)
 R. pseudosolanacearum (Phylotype III, GBBC_3272, Potato, Guinea)
 R. pseudosolanacearum (Phylotype III, NCPPB_1018, Potato, Angola)
 R. pseudosolanacearum (Phylotype III, NCPPB_332, Potato, Zimbabwe)
 R. syzygii (Phylotyp IV, Switzerland)
 R. syzygii subsp. syzygii (Phylotype IV, Clove, Indonesia)
 R. syzygii subsp. indonesiensis (Phylotype IV, Tomato, Indonesia)
 R. syzygii subsp. celebensis (Phylotype IV, Banana, Indonesia)
 R. syzygii (Phylotype IV, LMG_6976, Clover, Indonesia)
 R. pseudosolanacearum (Ginger, Switzerland)
 R. pseudosolanacearum (Ginger, Switzerland)

R. pseudosolanacearum (Ginger, Switzerland)	
Cross reaction with (Exclusivity)	None known
No cross reaction tested with (Exclusivity)	<i>Candidatus Arsenophonus phytopathogenicus</i> <i>Candidatus Phytoplasma solani</i> <i>Clavibacter michiganensis</i> subsp. <i>sepedonicus</i> <i>Pectobacterium parmentieri</i> / <i>P. wasabiae</i> <i>Pectobacterium atrosepticum</i> <i>Pectobacterium carotovorum</i> subsp. <i>Brasilense</i> <i>Pectobacterium carotovorum</i> subsp. <i>Carotovorum</i> <i>Dickeya solani</i> <i>Pseudomonas syringae</i> spp.
No matrix effect observed with (Selectivity)	<i>Capsicum annuum</i> (Bell pepper) - leaf and seed <i>Capsicum annuum</i> (chilli) - seed <i>Citrullus lanatus</i> (Watermelon) - leaf <i>Cucumis sativus</i> (Cucumber) – leaf <i>Musa</i> spp. (Banana) - leaf <i>Nicotiana tabacum</i> (Tobacco) – leaf <i>Rosa</i> spp. (Rose) - leaf <i>Solanum lycopersicum</i> (Tomato) – leaf and seed <i>Solanum melongena</i> (Eggplant) - leaf <i>Solanum tuberosum</i> (Potato) – tuber and leaf <i>Zingiber officinale</i> (Ginger) - rhizome

Sensitivity:

Diagnostic Sensitivity	100%
Analytical Sensitivity / LoD	10 ³ to 10 ⁴ CFU/ml
Sensitivity on host matrix	Potato tubers: 1 infected tuber in 299 healthy tubers (seed potato certification protocol): Ct ~21.
Other sensitivity characteristics	-

Validation:

Internal validation	2024
External validation	2025 (Belgium)
Reproducibility	100% (BIOREBA)
Repeatability	100% (BIOREBA)
Validation information	The internal validation was done with the internal collection of BIOREBA strains and species.

Validation release Date:
February 20th, 2025

QC manager:



Version: 2 – 20.02.2025