

Validation report

qPCR PLRV/PVY set/kits

Article No.: 839600 (qPCR PLRV/PVY set 96) / 839200 (qPCR PLRV/PVY set 192) /
839610 (qPCR PLRV/PVY kit 96/10) / 839625 (qPCR PLRV/PVY kit 96/25) /
839210 (qPCR PLRV/PVY kit 192/10) / 839225 (qPCR PLRV/PVY kit 192/25)

General information:

Target Pathogen	PLRV (Potato leafroll virus) / PVY (Potato virus Y)
Genus	<i>Polerovirus</i> (PLRV) / <i>Potyvirus</i> (PVY)
Method	Real-Time RT-qPCR, TaqMan (Multiplex)

Technical information:

Fluorophores	FAM: PVY Cy5: PLRV JOE(HEX): IC
Cycling program	<ul style="list-style-type: none"> ➤ 50°C for 20 min (Reverse Transcription) ➤ 95°C for 5 min (RT inactivation) ➤ 40 Cycles: <ul style="list-style-type: none"> ○ 95°C for 15 sec (Denaturation) ○ 60°C for 30 sec (Annealing / Extension)
Controls	Internal control (IC): artificial RNA included in Primers/Probes/IC Mix Negative control (NC): plant RNA Positive control (PC): RNA from PVY/PLRV-infected potatoes
Extraction	The qPCR PLRV/PVY kits include the potato DNA/RNA rapid extraction set, which is the validated extraction method for the qPCR PLRV/PVY set/kits.

Host matrix:

Tested plant material	Leaf, tuber (dormant), sprouts
Tested species infected	<i>Solanum tuberosum</i> (Potato) <i>Nicotiana tabacum</i> "Xanthi" (Tobacco) <i>Physalis floridana</i> (Bladder cherries)

Specificity:

Analytical Specificity	100%
Number of tested samples from target organisms (True Positives)	>100
Diagnostic Specificity	100%
Number of tested samples non-target organism (True Negatives)	>100
Detected isolates / geographic regions (Inclusivity)	<u>PLRV:</u> PLRV PV-0842 (Germany, Bladder cherries) PLRV W19-160 (Germany, Potato) PLRV 1 (Switzerland, Bladder cherries) PLRV Agata (Switzerland Potato) PLRV Valor 4 (Australia, Potato) PLRV PV-1212 (Germany, Bladder cherries) <u>PVY:</u> PVY W19-094 (Germany, Potato) PVY W19-095 (Germany, Potato)

	PVY PV-0343 / ON604831 (Germany, Potato) PVY N-605 (Switzerland, Tobacco) PVY N-605 Victoria (Switzerland, Potato) PVY O-803 (Switzerland, Tobacco) PVY O-768 1118 Victoria (Switzerland, Potato) PVY NTN 1317 Charlotte (Switzerland, Potato) PVY NTN 1214 Victoria (Switzerland, Potato) PVY W22-069 (Germany, Potato) PVY W22-063 (Germany, Potato)
Cross reaction with (Exclusivity)	None known
No cross reaction tested with (Exclusivity)	PVA (Potato virus A) PVM (Potato virus M) PVS (Potato virus S) PVX (Potato virus X)
No matrix effect observed with (Selectivity)	<i>Solanum tuberosum</i> (Potato) – Tuber and Leaf <i>Nicotiana tabacum</i> "Xanthi" (Tobacco) <i>Physalis floridana</i> (Bladder cherries)

Sensitivity:

Diagnostic Sensitivity	100%
Analytical Sensitivity / LoD	10^{-5} to 10^{-6}
Sensitivity on host matrix	Potato leaves: 10^{-5} to 10^{-6} Potato tubers: 10^{-5} to 10^{-6}
Other sensitivity characteristics	-

Validation:

Internal validation	2014/2015 + 2016/2017
External validation	2015 (Switzerland), 2017 (Switzerland) and proficiency test (PT) every year (Germany) since 2017.
Reproducibility	100%
Repeatability	100%
Validation information	Every year BIOREBA participates with PLRV/PVY qPCR kit on an external proficiency test (PT). The kit was in 2016/2017 validated (using 94 potato lots of 200 tubers each) for the testing in seed potato certification (comparison with DAS-ELISA, pools of 10 and 25 tubers are validated).

Validation release Date:
November, 7th, 2022

QC manager:



Version: 2 – 16.05.2024 – Adaptations to inclusivity and sensitivity information.