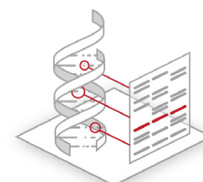


Validation Report: PCR Primers

PCR 94000 • Carmovirus-Pelarspovirus Group (Carmo-Pelarspo)



Test Characteristics

Test Name	Carmovirus-Pelarspovirus Group	Format	RT-PCR Primers
Catalog Number	94000	Extraction Method	Nucleic acid extraction
Acronym	Carmo-Pelarspo		
Genus	Carmovirus/Pelarspovirus		

Summary

The Carmovirus-Pelarspovirus Group PCR primers offer a sensitive diagnostic method to detect members of the Alphacarmovirus, Betacarmovirus, Gammacarmovirus, and Pelarspovirus genera of the Tombusviridae family. The primer sequences are based on conserved genome regions and can detect characterized and unassigned members of the Alphacarmovirus, Betacarmovirus, Gammacarmovirus, and Pelarspovirus genera.

Diagnostic Sensitivity

True Positives	1097
Correct Diagnoses	1097
Percent	100%

Analytical Specificity

Inclusivity:

Carmoviruses/Pelarspoviruses¹ Detected:

Virus Name	Species Name
Angelonia flower break virus (AnFBV)	Alphacarmovirus angeloniae
Calibrachoa mottle virus (CbMV)	Alphacarmovirus calibrachoe
Cardamine chlorotic fleck virus (CCFV)	Betacarmovirus cardaminis
Carnation mottle virus (CarMV)	Alphacarmovirus dianthi
Clematis chlorotic mottle virus (CICMV)	Pelarspovirus clematis
Cowpea mottle virus (CPMoV)	Gammacarmovirus vignae
Elderberry latent virus (ELV)	Pelarspovirus sambuci
Hibiscus chlorotic ringspot virus (HCRSV)	Betacarmovirus hibisci
Honeysuckle ringspot virus (HRSV) ²	Alphacarmovirus lonicerae
Japanese iris necrotic ring virus (JINRV) ²	Betacarmovirus iridis
Melon necrotic spot virus (MNSV)	Gammacarmovirus melonis
Nootka lupine vein clearing virus (NLVCV) ²	Alphacarmovirus lupini
Pea stem necrosis virus (PSNV) ²	Gammacarmovirus pisi
Pelargonium chlorotic ring pattern virus (PCRPV)	Pelarspovirus chloropelargonii



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Carmoviruses/Pelarspoviruses¹ Detected:

Virus Name	Species Name
Pelargonium flower break virus (PFBV)	Alphacarmovirus pelargonii
Pelargonium line pattern virus (PLPV)	Pelarspovirus lineapelargonii
Pelargonium ringspot virus (PelRSV)	Pelarspovirus anulopelargonii
Rosa rugosa leaf distortion virus (RRLDV) ²	Pelarspovirus rosae
Saguaro cactus virus (SgCV)	Alphacarmovirus cacti
Soybean yellow mottle mosaic virus (SYMMV) ²	Gammacarmovirus glycinis
Turnip crinkle virus (TCV)	Betacarmovirus brassicae
¹ The list above represents viruses that have been shown to be detected by this group PCR test. It also represents viruses that may be detected based on <i>in silico</i> analysis. If you have confirmed detection of a predicted virus detection or a virus not on this list, please contact us. We would like to work with you to further validate detection capabilities.	
² Predicted detection by <i>in silico</i> analysis only	

Carmoviruses/Pelarspoviruses Not Detected:

Virus Name	Species Name
None Known	

Exclusivity:**Cross-reacts With:**

Virus Name	Species Name
Galinsoga mosaic virus (GAMV) ¹	Gallantivirus galinsogae
Maize chlorotic mottle virus (MCMV)	Machlomovirus zeae
Tobacco necrosis virus A (TNV-A)	Alphanecrovirus nicotianae
Trailing lespedeza virus 1 (TLV1) ¹	Tralespevirus lespedezae
¹ Predicted detection by <i>in silico</i> analysis only	

Does Not Cross-react With:

Virus Name	Species Name
None Known	

Diagnostic Specificity

True Negatives 12159
Correct Diagnoses 12159
Percent 100%

Selectivity:**No Matrix Effect Observed With:**

Achillea leaves	Agapanthus leaves	Alstroemeria leaves	Angelonia leaves
Aster leaves	Astilbe leaves	Astilbe roots	Buddleia leaves
Carnation leaves	Clematis leaves	Colocasia leaves	Colocasia roots
Coreopsis leaves	Crisantemo leaves	Dahlia leaves	Delphinium leaves
Dianthus leaves	Dieffenbachia leaves	Echinacea leaves	Epimedium leaves



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Gaillardia leaves	Geranium leaves	Gypsophila leaves	Hemp leaves
Hibiscus leaves	Leucanthemum leaves	Mandevilla leaves	Melon seeds
Osteospermum leaves	Palm leaves	Pelargonium leaves	Phlox leaves
Scabiosa leaves	Schlumbergera leaves	Sedum leaves	Syngonium leaves
Syngonium roots	Thymus leaves	Tobacco leaves	Tradescantia leaves
Verbena leaves	Veronica leaves		
The hosts on the above list have been chosen to represent those which historically cause a range of matrix effects, in addition to those expected to be screened for this pathogen. Not all plant species susceptible to this pathogen have been screened, but may still be used with this assay unless otherwise noted below. As with all diagnostic tools, Agdia recommends confirming all results with a secondary detection method before making any economic decisions (ex: discarding plants due to positive test results, etc.).			

Matrix Effect Observed With:			
None Known			

Robustness

Planned deviation analysis:

No deviations from the user guide protocol were validated.

Stability:

	1-year stability (accelerated)	Real-time Stability Verification
Positive Sample (High)	Pass	Monitoring
Positive Sample (High)	Pass	Monitoring
Positive Sample (Low)	Pass	Monitoring
Positive Sample (Low)	Pass	Monitoring
Positive Sample (Low)	Pass	Monitoring
Negative Sample	Pass	Monitoring
Negative Sample	Pass	Monitoring
Negative Sample	Pass	Monitoring



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Glossary

Diagnostic sensitivity¹:	The percentage of positive samples correctly identified in an experiment with known positive controls.
Diagnostic specificity¹:	The percentage of negative samples correctly identified in an experiment with known negative controls.
Analytical sensitivity²:	The smallest amount of target that can be detected reliably (this is sometimes referred to as the 'limit of detection')
Analytical specificity²:	(comprises inclusivity and exclusivity)
Inclusivity³:	The performance of a test with a range of target isolates covering genetic diversity, different geographical origin and/or hosts associated with the target organism.
Exclusivity³:	The performance of a test with a range of non-targets (e.g. cross-reaction with closely related organisms, contaminants)
Selectivity²:	The level of effect that matrices and relevant plant parts have on the performance of the assay.
Repeatability²:	The agreement between test replicates of the same sample tested by the same operator.
Reproducibility³:	The ability of a test to provide consistent results when applied to aliquots of the same sample tested under different conditions (e.g. time, users, equipment, location)
Robustness^{1,3}:	The extent to which varying test conditions (e.g. temperature, volume, change of buffers) affect the established test performance values. May also be referred to as planned deviation analysis.
Stability¹:	The performance of test reagents or controls over time.

References:

¹Groth-Helms, D., Rivera, Y., Martin, F. N., Arif, M., Sharma, P., Castlebury, L. A. (in press). Terminology and Guidelines for Diagnostic Assay Development and Validation: Best Practices for Molecular Tests. *PhytoFrontiers*.

²Eads, A., Groth-Helms, D., Davenport, B., Cha, X., Li, R., Walsh, C., Schuetz, K., (in press). The Commercial Validation of Three Tomato Brown Rugose Fruit Virus Assays. *PhytoFrontiers*.

³EPPO (2018) PM 7/76 (5) Use of EPPO Diagnostic Standards, EPPO Bulletin 48, 373– 377.



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