

ImmunoStrip® Validation Report On-site Plant Pathogen Testing *Capsicum chlorosis virus* (CaCV) ISK/STX 62800 ImmunoStrip®

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Test Characteristics

Test Name	Capsicum chlorosis virus	Capture Antibody	Polyclonal (Rabbit)
Catalog Number	62800	Detection Antibody	Polyclonal (Rabbit)
Acronym	CaCV	Format	Lateral Flow Device
Genus	Orthotospovirus	Diluents	SEB1
Binomial Name	Orthotospovirus capsiciflavi	Sample Dilution	1:20

Summary

The Capsicum chlorosis virus (CaCV) ImmunoStrip[®] is used to detect the presence of CaCV in ornamental and vegetable crops. ImmunoStrips[®] are the perfect screening tool for use in the field, greenhouse, and the lab.

Diagnostic Sensitivity		Analytical Sensitivity		
True Positives	27	Limit of Detection:	1:256 dilution of infected tissue (pathogen titer unknown)	
Correct Diagnoses	27			
Percent	100%			

Analytical Specificity

Inclusivity:

This assay was designed to detect all strains and isolates of CaCV. Twenty-seven distinct samples of CaCV have been experimentally proven to be detected including samples of CaCV-PV-0864 from Thailand and a Grecian isolate.

Exclusivity:

Cross-reacts With:	
None Known	

Does Not Cross-react With:

Alfamovirus AMV (Alfalfa mosaic virus) (AMV)	Cucumovirus CMV (Cucumber mosaic virus) (CMV)
Nepovirus lycopersici (Tomato ringspot virus) (ToRSV)	Nepovirus nicotianae (Tobacco ringspot virus) (TRSV)
Orthotospovirus arachinecrosis (Groundnut bud necrosis virus) (GBNV)	Orthotospovirus citrullomaculosi (Watermelon silver mottle virus) (WSMoV)
Orthotospovirus glycininecrovenae (Soybean vein necrosis virus) (SVNV)	Orthotospovirus impatiensnecromaculae (Impatiens necrotic spot virus) (INSV)
Orthotospovirus iridimaculaflavi (Iris yellow spot virus) (IYSV)	Orthotospovirus tomatoflavi (Tomato chlorotic spot virus) (TCSV)
Orthotospovirus tomatomaculae (Tomato spotted wilt virus) (TSWV)	Potexvirus cymbidii (Cymbidium mosaic virus) (CymMV)
Potexvirus ecspotati (Potato virus X) (PVX)	Potexvirus pepini (Pepino mosaic virus) (PepMV)
Tobamovirus fructirugosum (Tomato brown rugose fruit virus) (ToBRFV)	Tobamovirus odontoglossi (Odontoglossum ringspot virus) (ORSV)
Tobamovirus tabaci (Tobacco mosaic virus) (TMV)	Tobamovirus tomatotessellati (Tomato mosaic virus) (ToMV)
Tombusvirus cymbidii (Cymbidium ringspot virus) (CymRSV)	Tombusvirus lycopersici (Tomato bushy stunt virus) (TBSV)

Diagnostic Specificity

True Negatives 86 Correct Diagnoses 86

Percent 100%

Selectivity:

No Matrix Effect Observed With:			
Cattleya leaves	Citrullis leaves	Cucumber leaves	Cymbidium leaves
Dendrobium leaves	Gloxinia leaves	Haemanthus leaves	Hippeastrum leaves
Hoya leaves	Odontoglossum leaves	Peanut leaves	Pepper fruit
Pepper leaves	Phalaenopsis leaves	Pineapple leaves	Rudbeckia leaves
Spathoglottis leaves	Tomato fruit	Tomato leaves	Zantedeschia 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			

Repeatability

Reproducibility

- Number of Samples 32
- Replicates per Sample 3
- Number of Operators 4
 - Total Replicates 384
- Replicates in Agreement 384
 - Percent Agreement 100%

Robustness

Planned deviation analysis:

No deviations from the user guide protocol were validated.

Number of Samples 111

Replicates per Sample 2 - 3

Replicates in Agreement 324 Percent Agreement 100%

Total Replicates 324

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

Glossary

Diagnostic sensitivity ¹ :	The percentage of positive samples correctly identified in an experiment with known positive controls.
Diagnostic specificity ¹ :	The percentage of negative samples correctly identified in an experiment with known negative controls.
Analytical sensitivity ³ :	The smallest amount of target that can be detected reliably (this is sometimes referred to as the 'limit of detection')
Analytical specificity ³ :	(comprises inclusivity and exclusivity)
Inclusivity ³ :	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.
Exclusivity ³ :	The performance of a test with a range of non-targets (e.g. cross-reaction with closely related organisms, contaminants)
Selectivity ² :	The level of effect that matrices and relevant plant parts have on the performance of the assay.
Repeatability ² :	The agreement between test replicates of the same sample tested by the same operator.
Reproducibility ³ :	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.
Stability ¹ :	The performance of test reagents or controls over time.
Poforoncos:	

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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