AmplifyRP® XRT for HSVd
Validation Report
Hop stunt viroid
Product No. XCS 64200

Test Characteristics

Test Name	Hop stunt viroid	**Test Label**	FAM-labeled target probe
Catalog Number	64200	**Internal Control**	N/A
Acronym	HSVd	**Format**	XRT
Genus	Hostuviroid	**Diluents**	GEB/PD1
		Sample Dilution	1:20

Summary

AmplifyRP XRT for HSVd is a rapid RNA amplification and detection platform designed for field-based or laboratory testing of hop for Hop stunt viroid. This kit includes lyophilized reaction pellets containing the necessary reagents to amplify HSVd RNA at a single operating temperature (42 °C).

Diagnostic Sensitivity

True Positives	22
Correct Diagnoses	22
Percent	100%

Analytical Sensitivity

Limit of Detection: The assay has a 100% detection rate at 10 fg/µL with RNA transcripts. (n=2)

Analytical Specificity

Inclusivity:

Isolates and Geographic Regions Detected:

HSVd citrus-type	HSVd hop-type
HSVd plum-type	

Exclusivity:

Cross-reacts With:

None Known	

Does Not Cross-react With:

Apple fruit crinkle viroid (AFCVd)[1]	Apple mosaic virus (ApMV)
Apricot latent virus (ApLV)	Chrysanthemum stunt viroid (CSVd)
Citrus exocortis viroid (CEVd)	Columnea latent viroid (CLVd)
Hop latent viroid (HLVd)	Hop latent virus (HpLV)
Nepovirus arabis (Arabis mosaic virus) (ArMV)	Pepper chat fruit viroid (PCFVd)
Potato spindle tuber viroid (PSTVd)	Tomato apical stunt viroid (TASVd)
Tomato chlorotic dwarf viroid (TCDVd)	Tomato planta macho viroid (TPMVd)
[1]AFCVd is a tentative member of the genus Apscaviroid	

Diagnostic Specificity

True Negatives	45
Correct Diagnoses	45
Percent	100%

Selectivity:

No Matrix Effect Observed With:			
Calibrachoa leaves	Citrus leaves	Grape leaves	Hop leaves
Plum leaves	Tomato leaves		

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
Positive Sample (Low)	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.

References:

[1]Groth-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.
[2]Eads, 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.
[3]EPPO (2018) PM 7/76 (5) Use of EPPO Diagnostic Standards, EPPO Bulletin 48, 373– 377.

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