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Supplementary Materials: The p22 RNA Silencing Suppressor of the Crinivirus Tomato chlorosis virus is Dispensable for Local Viral Replication but Important for Counteracting an Antiviral RDR6-Mediated Response during Systemic Infection

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Table S1. Nucleotide sequences of primers used in this work.

Gene/cDNA Clone	Primer		Primer Sequence (5'-3') 1	Primer Position 1,2
ToCV-RNA1 clone	MA 1543	(FW)	AA <u>CTGCAG</u> AAGCCATTGACA	6907–6926
	MA 1544	(RV)	TTGAATATTAACCAGA/ACTGACCTAAATAACAACAGTC	8259-8244/7661-7640
	MA 1545	(FW)	ATTTAGGTCAGT/TCTGGTTAATATTCAAAGGATGG	7650-7661/8244-8266
	MA 1546	(RV)	GAT <u>CTGCAG</u> GTCGACTCTAG	8620-8601
ToCV-RNA2 clone	MA 1178	(FW)	ACCGGGCGCAGTTCATACAA	1522–1541
	MA 1179	(RV)	CCGACAAGAAACAGCGCTCC	1697–1675
CAC	MA 1279	(FW)	CTCCGTTGTGATGTAACTGG	779–798
	MA 1280	(RV)	ATTGGTGGAAAGTAACATCATC	950–929

¹ The underlined sequences are restriction endonucleases sites introduced into the primers for cloning (*Pst* I in MA 1543 and MA 1546). Nucleotides in bold indicate overlapping sequences. Nucleotide sequences in italics and positions separated with the slash symbol (/) indicate the p22 flanking sequences. FW and RV indicate forward and reverse sense primers, respectively; ² Primer positions indicate the positions in the RNA1 and RNA2 nucleotide sequences of AT80/99 ToCV isolate (GenBank accession numbers DQ983480 and DQ136146, respectively) and the CAC gene of Solanaceae (GenBank accession number XM_009783969.1) used as internal standard.

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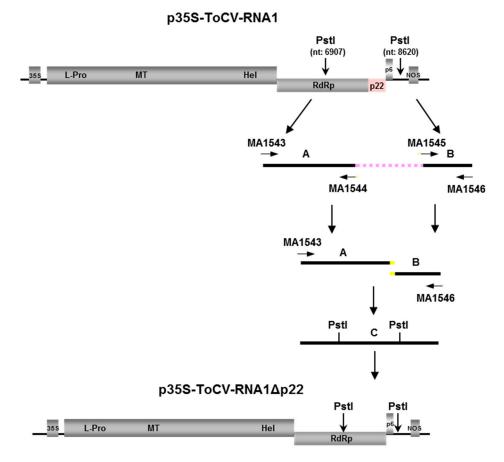


Figure S1: Diagram of the methodology used to create the p22-deficient ToCV RNA1 mutant clone.

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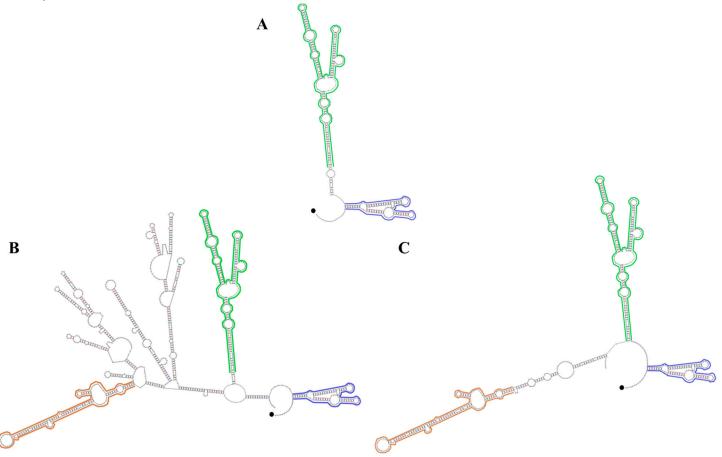


Figure S2. Lowest free energy secondary structure prediction of 3'-ends of the coding strand of ToCV RNA1. (**A**) Predicted secondary structure of the entire 3'-UTR (nt: 8237-8594); (**B**) Predicted secondary structure of the 3'-end comprising the 3'-UTR, the p22 sequence and the 241 nt located before the p22 sequence (nt: 7421-8594); (**C**) Predicted secondary structure of the 3'-end comprising the 3'-UTR and the 241 nt located before the p22 sequence (nt: 7421-7662 and 8237-8594). Identical stem loops are highlighted with similar colors. The 3'-end of each structure is indicated with (•). The RNAs were folded using the mfold program (Zuker, 2003).



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