

Supporting Information

Foliar Delivery of siRNA Particles for Treating Viral Infections in Agricultural Grapevines

Aviram Avital^{1,2,†}, Noy Sadot Muzika^{3,†}, Zohar Persky^{3,†}, Avishai Karny¹, Gili Bar¹, Yuval Michaeli¹, Jeny Shklover¹, Janna Shainsky¹, Haim Weissman^{4,}, Oded Shoseyov,^{3,*} Avi Schroeder^{1,*}*

Figure S1: Sequence design for RdRp

5'- GTTAGCGGTGATGATAGCCTTATATTTAGTCGGCAGCCGTTGGATATTGATACGTCGGTT
CTGAGCGATAATTTTGGTTTTGACGTAAAGATTTTAAACCAAGCTGCTCCATATTTTGT
TCTAAGTTTTTAGTTCAAGTCGAGGATAGTCTCTTTTTTGTTCCTCCGATCCACTTAAACTC
TTCGTTAAGTTTGGAGCTTCCAAAACCTTCAGATATCGACCTTTTACATGAGATTTTTCAA
TCTTTCGTCG-3'

Figure S2: Sequence design for CP

5'-CCAGCGCAAGTGGCGGAACACAGGAAACCGATATAGGGGTAGTGCCGGAATCTGA
GACTCTCACACCAAATAAGTTGGTTTTCGAGAAAGATCCAGACAAGTTCTTGAAGACT
ATGGGCAAGGGAATAGCTTTGGACTTGGCGGGAGTTACCCACAAACCGAAAGTTATT
AACGAGCCAGGGAAAGTATCAGTAGAGGTGGCAATGAAGATTAATGCCGCATTGATG
GAGCTGTGTAAGAAGGTTATGG-3'

Figure S3: dsRNA-ImPEI encapsulation efficiency

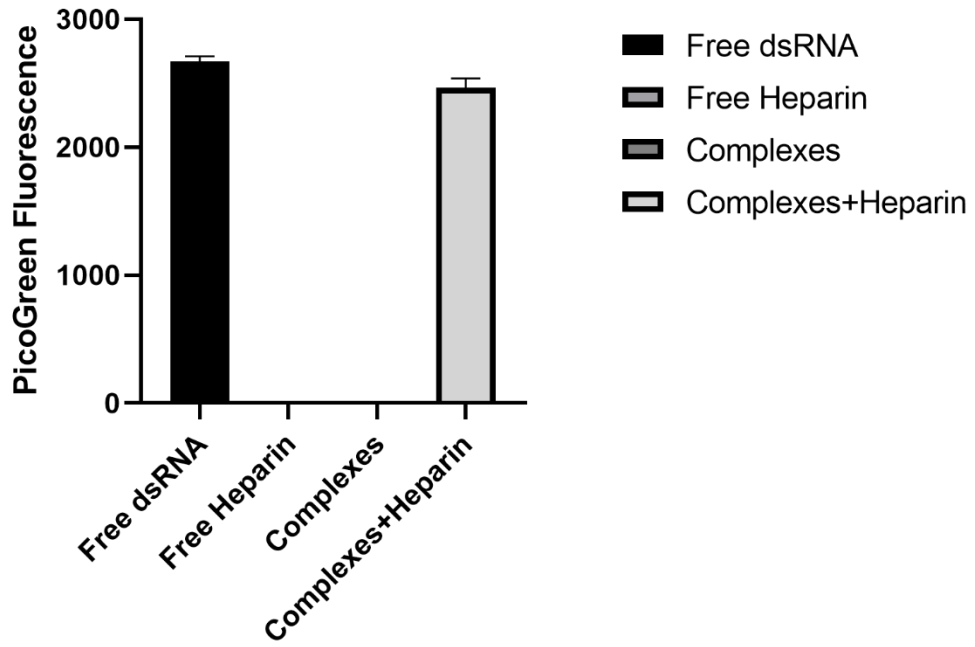
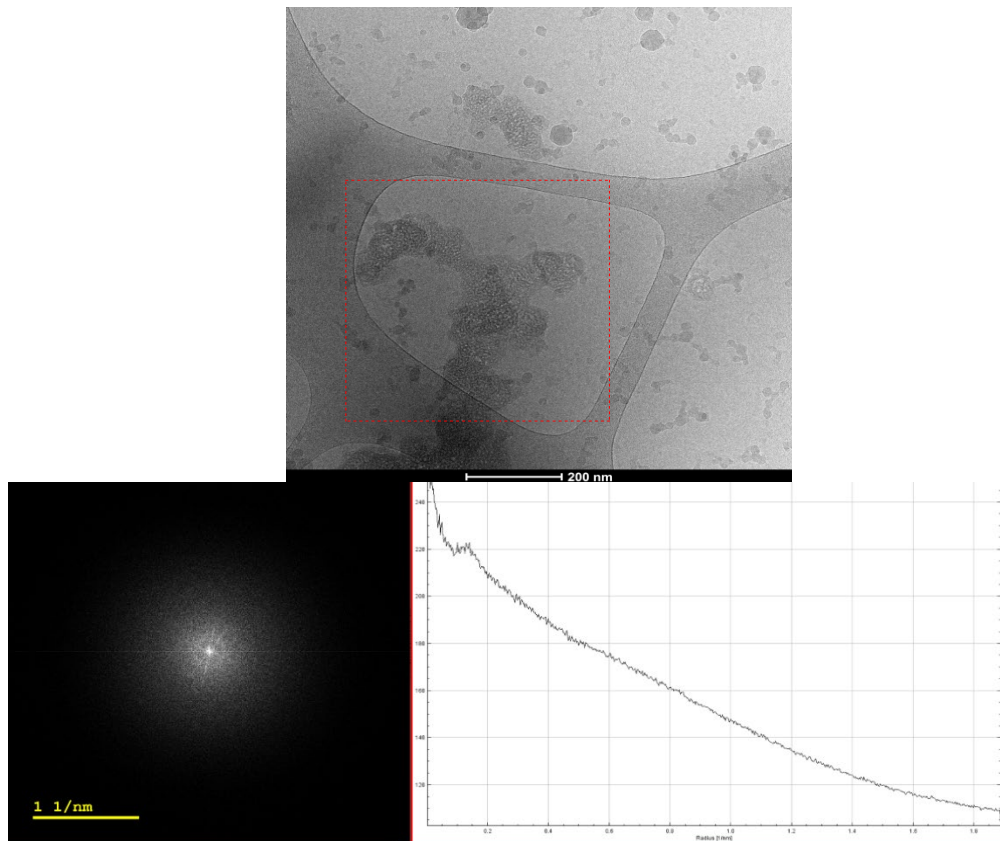


Figure S4: Additional FFT and radial integration

A



B

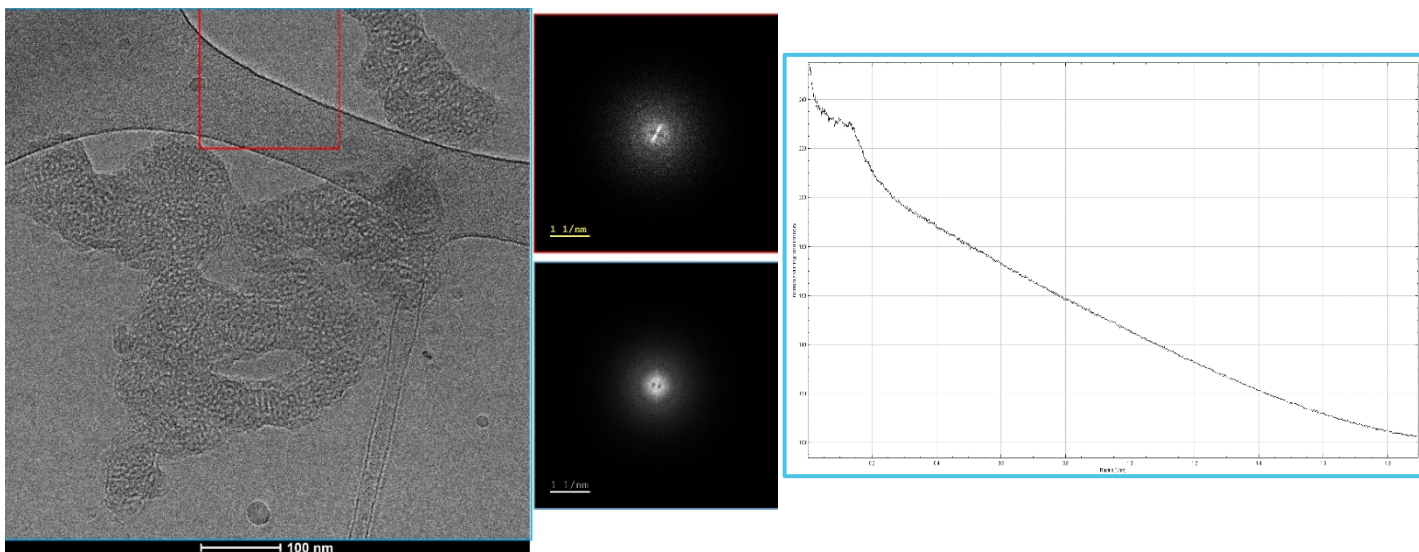
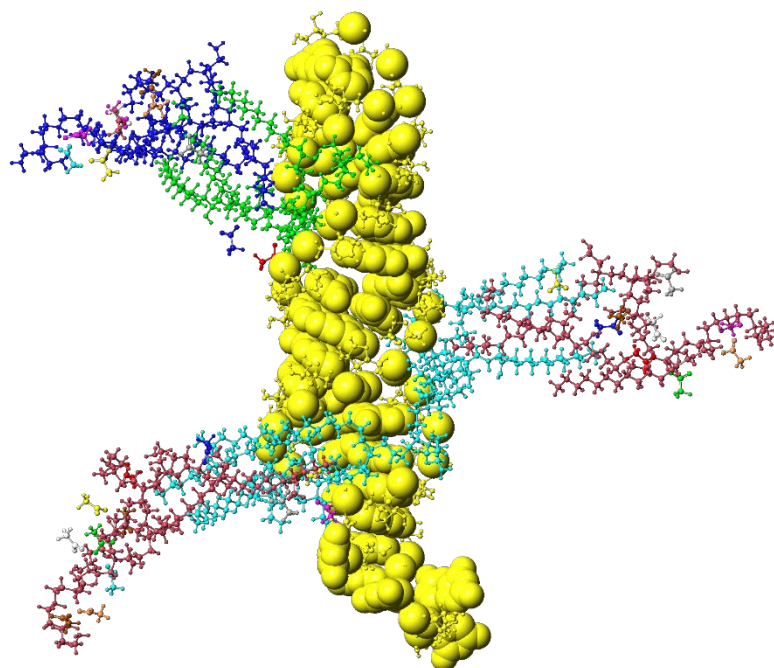


Figure S5: dsRNA-ImPEI partially energy minimized molecular mechanics model



L

Figure S6: Free Cy5 biodistribution

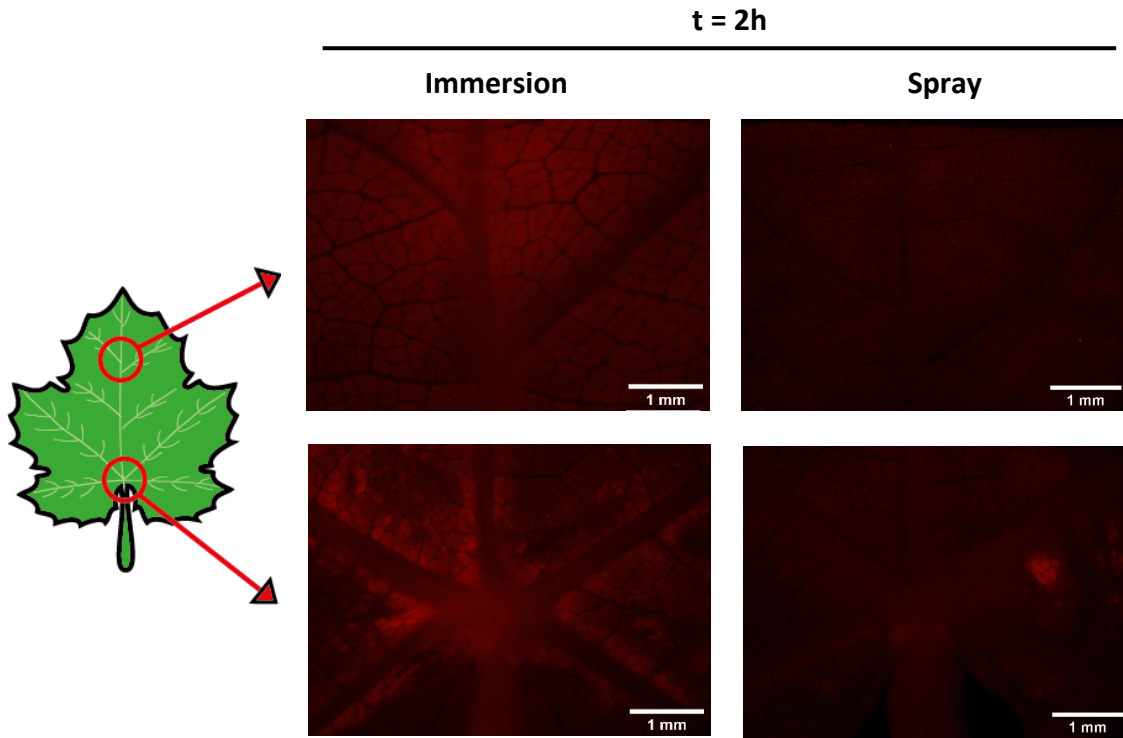


Figure S7: Heparin release assay

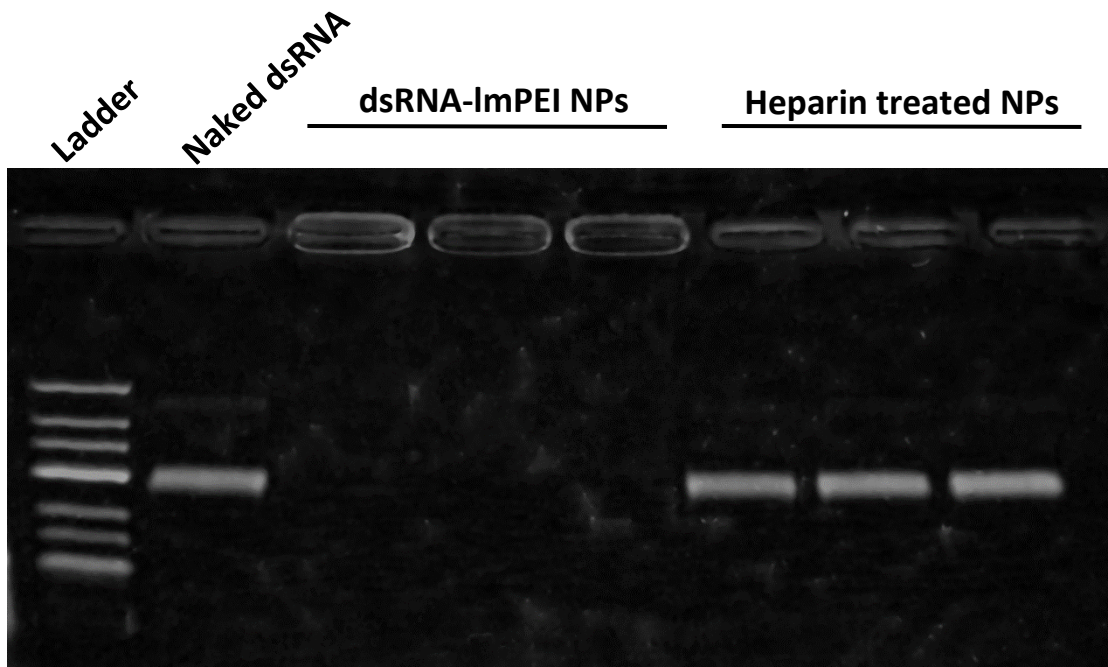


Figure S8: GLRaV3 relative expression in 2019 field experiment

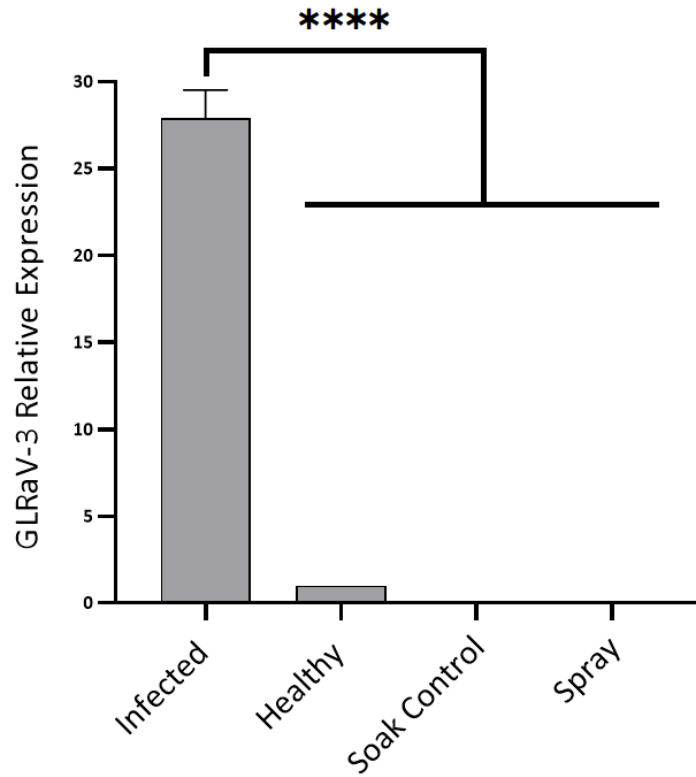


Figure S9: GLRaV3 relative expression throughout 2020 field experiment

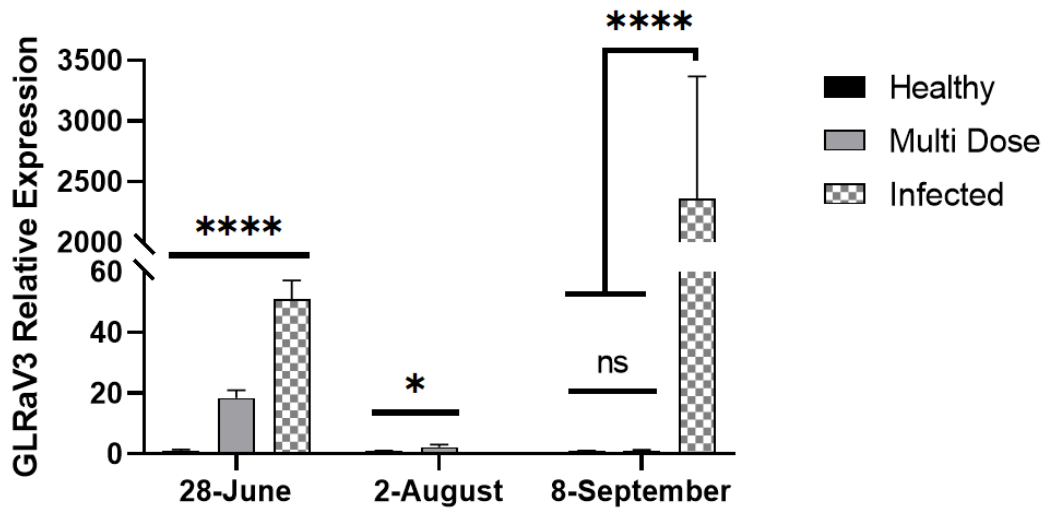


Figure S10: pH after 3, 5 and 8 weeks post treatment in 2019 experiment

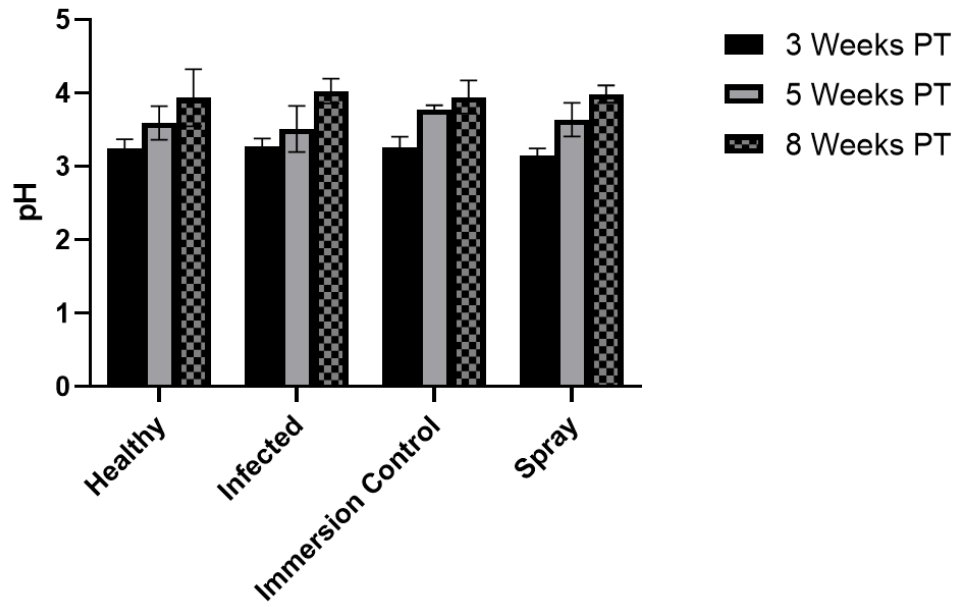


Figure S11: Tannin index after 3, 5 and 8 weeks post treatment in 2019 experiment

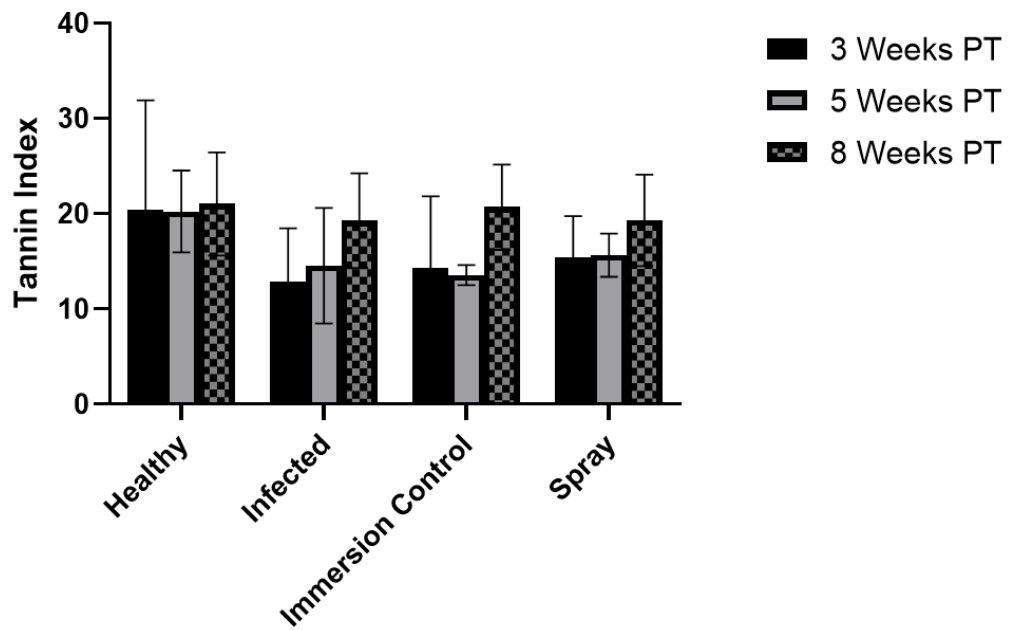


Figure S12: Total acid after 3, 5 and 8 weeks post treatment in 2019 experiment

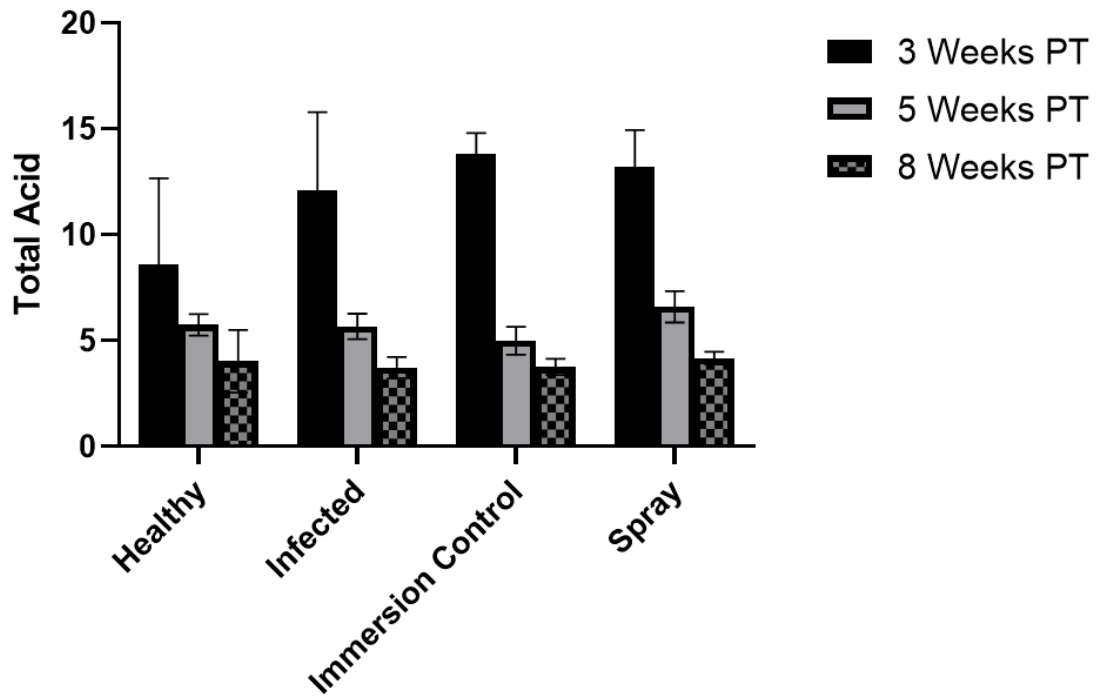


Figure S13: Color density after 3, 5 and 8 weeks post treatment in 2019 experiment

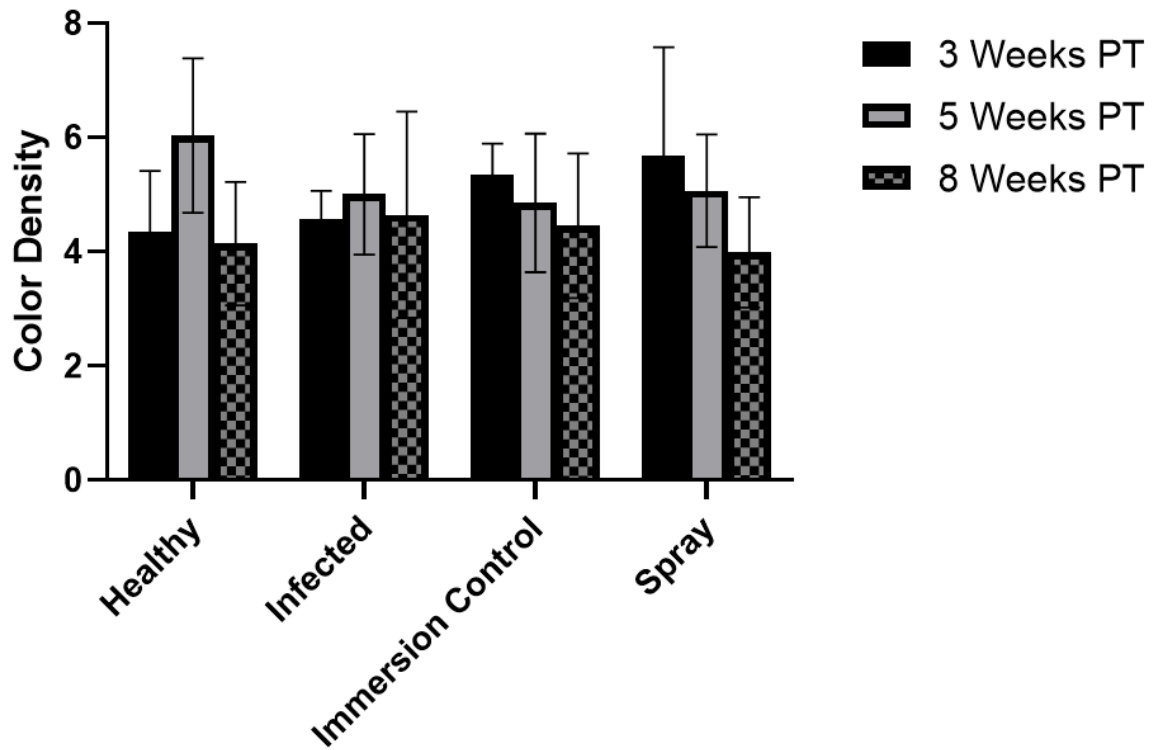


Figure S14: Softness ratio after 3, 5 and 8 weeks post treatment in 2019 experiment

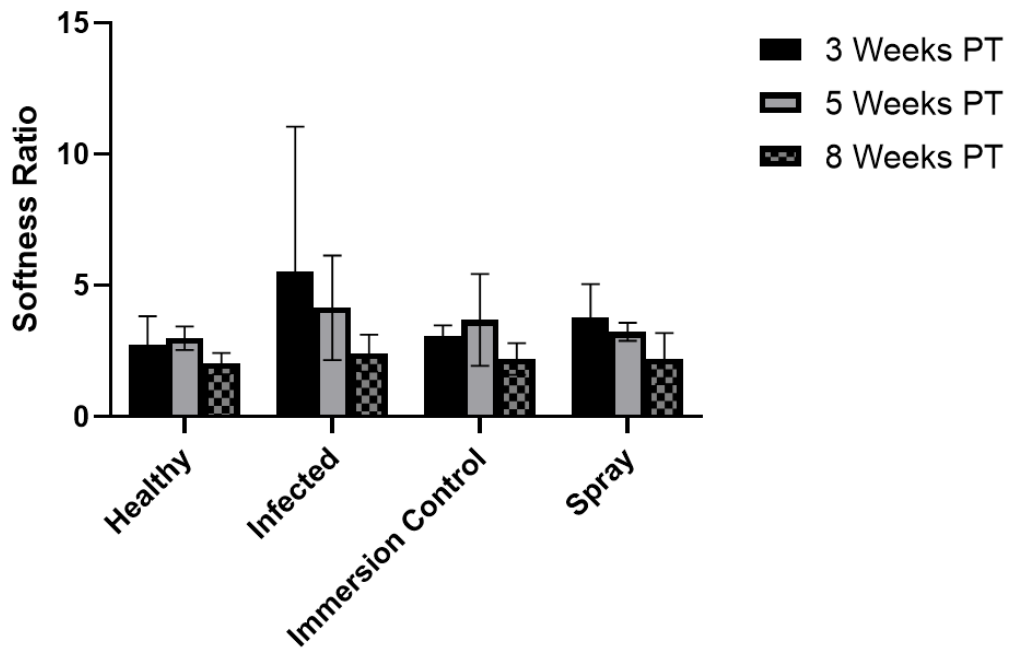







Table S1. Forward and reverse primers for GLRaV strains identification

Primer Name	Sequence 5' → 3'
GLRAV-1_F	CTAGCGTTATATCTCAAATGA
GLRAV-1_R	CCCATCACTTCAGCACATAAA
GLRAV-2_F	TTGACAGCAGCCGATTAAGCG
GLRAV-2_R	CTGACATTATTGGTGCGACGG
GLRAV-3_F	CGCTAGGGCTGTGGAAGTATT
GLRAV-3_R	GTTGTCCCGGTACCAGATAT
GLRAV-4_F	ACATTCTCCACCTTGTGCTTT
GLRAV-4_R	CATACAAGCGAGTGCAATTACA
GLRAV-7_F	TATATCCCAACGGAGATGGC
GLRAV-7_R	ATGTTCTCCACCAAATCG
GLRAV-9_F	CGGCATAAGAAAAGATGGCAC
GLRAV-9_R	TCATTCACCACTGCTTGAAC

Table 2. Primer sets used in this study and their amplification efficiencies [26]

Primer Name	Sequence 5' → 3'	Primer Efficiency
<i>Vitis Vinifera</i> Actin	Forward: CTTGCATCCCTCAGCACCTT	92.235%
	Reverse: TCCTGTGGACAATGGATGGA	
<i>Vitis Vinifera</i> GAPDH	Forward: TTTGGTATTAGGAACCCAGAGGA	99.602%
	Reverse: CAACAACGAACATAGGAGCATCTT	
<i>Vitis Vinifera</i> Ef1- α	Forward: GAACTGGGTGCTTGATAGGC	96.0%
	Reverse: AACCAAAATATCCGGAGTAAAAGA	
<i>Grapevine leafroll-associated virus 3</i> Hsp70	Forward: GCYTGTGGRGCTAAGGTTTAC	102.432%
	Reverse: ARCAGGGTATTGGACTRCCTTTTCG	

Table S3. Infection severity scoring

Score	0	1	2	3	4
Visual description					
Literal description	Green leaves without any visible symptoms	Appearance of reddish stains in the interveinal areas of mature leaves near the basal part of the shoots	Symptoms extend upward to other leaves and stains expand	Only a narrow part on either side of the main veins remains green	Leaves are completely red and margins are curled inwards