

Supplementary Table 1. Primer list used in this study

Assay	Primer	Sequence 5'→3'	Target gene	Size (bp)	Reference
RT-PCR	HSVd-F	GCC CCG GGG CTC CTT TCT CAG GTA AG	HSVd	303	Astruc et al. (1996)
	HSVd-R	CGC CCG GGG CAA CTC TTC TCA GAA TCC			
	Nad5-F	GAT GCT TCT TGG GGC TTC TTG TT	Internal control	181	Menzel et al. (2002)
	Nad5-R	CTC CAG TCA CCA ACA TTG GCA TAA			
RT-dPCR	HSVd-F	CAT CAC CTC TCG GTT CGT CT	HSVd	101	In this study
	HSVd-P	6-FAM-CGC GGA TCC TCT CTT GAG CCC-BHQ-1			
	HSVd-R	WTG CCG CAA CTC GAG AAT			

RT-PCR, reverse transcription polymerase chain reaction; HSVd, hop stunt viroid.

Supplementary Table 2. Data summary from Oxford nanopore

Sample name	Raw reads			
	Total reads	Minimum read length	Maximum read length	Mean read length
188.08	336,727	66	7438	330.9

Supplementary Table 3. Comparison of the growth of grapevine plantlets treated with garlic extract and DEPC

	Length of shoot	Length of root
Control 1	6.2	10
Control 2	12.5	8.4
Control 3	12.4	11.9
1	13.5	18
2	15	18.2
3	14	15.4
4	15.5	9.2
5	16.3	9.6
6	15.5	12
7	14	14
8	14.7	17.7
9	14.7	19.8
10	14.9	21.6
11	14.6	18
12	15.4	14
13	17	13.6
14	16.4	11
15	13	15.6
16	14.1	11.4
17	14.5	17
18	25.9	11.8
19	16.5	11.1
20	14.3	13
21	15	11.9

Supplementary Table 4. Detection of HSVd in samples of grapevine plantlets by RT-PCR

	Control 1	Control 2	Control 3	1	2	3	4	5	6	7	8	9	10	11	12	13	14
RT-PCR (28 dpt)	+	+	+	-	-	-	-	-	-	+	-	-	-	-	-	-	-
	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31
RT-PCR (28 dpt)	-	-	-	-	+	-	-	-	-	-	+	-	-	-	-	-	-
	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46	47	48
RT-PCR (28 dpt)	-	-	-	-	-	-	+	+	+	+	-	+	-	+	+	+	+
	49	50	51	52	53	54	55	56	57	58	59	60	61	62	63	64	65
RT-PCR (28 dpt)	+	+	-	-	-	-	-	-	-	+	-	-	-	+	+	+	+
	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82
RT-PCR (28 dpt)	-	+	-	+	-	-	-	-	-	-	-	-	-	-	-	-	+
	83	84	85	86	87	88	89	90	91	92	93	94	95	96	97	98	99
RT-PCR (28 dpt)	-	+	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-
Inhibition rate	74.45% (74/99)																

HSVd, hop stunt viroid; RT-PCR, reverse transcription polymerase chain reaction; dpt, days post-treatment.

Supplementary Table 5. Concentration of HSVd in micropropagated grapevine plantlets treated with garlic extract by dPCR

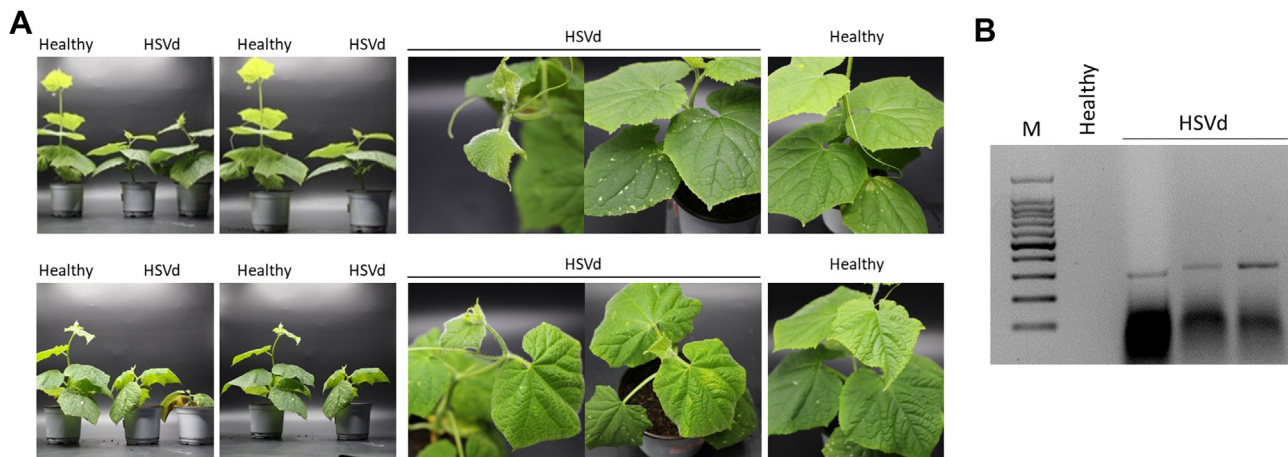
Sample name	Concentration (copies/ μ l)		Inhibition rate (%)	Sample name	Concentration (copies/ μ l)		Inhibition rate (%)
	0 dpt	28 dpt			0 dpt	28 dpt	
Control	10,662.7	10,658.6	0.04	17	12,352.6	1.865	99.9
1	9,549.6	4,147.8	56.6	18	12,349	956.4	92.2
2	9,549.1	4,887.9	48.8	19	12,351.8	66.54	99.4
3	19,528	931.3	95.2	20	5,499.4	808.3	85.3
4	27,475	2,147.6	92.2	21	5,469.9	617.5	88.7
5	3,960.9	801.4	79.8	22	8,762.9	957.4	89.0
6	26,194	564.5	97.8	23	9,054.4	501.3	94.4
7	38,410	2,009.5	94.8	24	2,012.8	254.5	87.3
8	3,226.2	573.4	82.2	25	56,993	1668.8	97.0
9	17,296	458.9	97.3	26	12,351	0	100
10	12,350.9	2,104.1	83.0	27	9,820	0	100
11	3,263.9	591.6	81.9	28	8,830.7	0	100
12	6,406.5	1,200.8	81.3	29	5,585.3	125.7	97.7
13	5,876	0.766	99.9	30	12,351.2	414.2	96.6
14	4,047.3	858.3	78.8	31	12,349.1	0.049	99.9
15	7,709	230.5	97.0	32	6,987	0	100
16	2,462.1	1,443.7	41.4	33	5,699.3	1297.3	77.2

HSVd, hop stunt viroid; dpt, days post-treatment.

Supplementary Table 6. Concentration of HSVd in micropropagated grapevine plantlets treated with garlic extract by dPCR

Sample name	Concentration (copies/ μ l)		
	0 dpt	2nd generation	4th generation
Control	12,349.9	3,960.2	4,478.8
1	42,504	116.0	0
2	42,362	0	0
3	35,971	745.4	298.3
4	53,652	0	0
5	12,351	0	0
6	9,820	0	0
7	8,830.7	0	0
8	6,987	0	0
9	37,391	0	0

HSVd, hop stunt viroid; dPCR, digital PCR.



Supplementary Fig. 1. Pathogenicity test of hop stunt viroid (HSVd) in cucumber plants. (A) Visual symptoms and growth comparison of HSVd-inoculated cucumber plants at 25 days post-inoculation. Healthy: untreated plants; HSVd: plants inoculated HSVd. (B) Detection of the HSVd in cucumber plants inoculated HSVd by reverse transcription polymerase chain reaction.

References

- Astruc, N., Marcos, J. F., Macquaire, G., Candresse, T. and Pallás, V. 1996. Studies on the diagnosis of hop stunt viroid in fruit trees: identification of new hosts and application of a nucleic acid extraction procedure based on non-organic solvents. *Eur. J. Plant Pathol.* 102:837-846.
- Menzel, W., Jelkmann, W. and Maiss, E. 2002. Detection of four apple viruses by multiplex RT-PCR assays with coamplification of plant mRNA as internal control. *J. Virol. Methods* 99:81-92.