

Supplementary Information

Analyses of virus/viroid communities in nectarine trees by next-generation sequencing and insight into viral synergisms implication in host disease symptoms

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Supplementary Data

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Table S1 sRNA sequencing, contig assembly, and virus/viroid genome identification.

Table S2 Oligonucleotide primers used for amplification of virus and viroid sequences in this study.

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Table S1. sRNA sequencing, contig assembly, and virus/viroid genome identification.

Sample	Number of raw sRNA reads	Clean sRNA reads	Mapped sRNA reads	Mapped sRNA/ Clean reads	Number of contigs	Contig length (nt)
T01	22434184	20906239	1156297	5.53%	687	33-366
T02	23559998	22191401	2129850	9.60%	744	33-338
T03	30438485	28435618	316928	1.11%	52	33-474
T04	29588745	24849851	672912	2.71%	243	33-408
T05	27056398	25000656	566883	2.27%	15	33-84

Table S2 Oligonucleotide primers used for amplification of virus/viroid sequences.

Primer		Primer sequence (5'-3')	Position ^a	Product size (bp)	References ^b
APV1	F	ATGAGTACCTCTTCTGCAACAG	CP	1210	This study
	R	ACCCTCAATCCATCTCCTTCC			
APV2	F	AGGTCCAGTGTGTGTAGATTGA	CP	1235	This study
	R	CAACAACAGTCTTGACCGGG			
APV3	F1	ACTGGTGAGTCTGTGGTAGT	CP	1389	This study
	R1	TGGGCGACTAATCCATCTCC			
	F2	ACTCTACAGGTACATTCGGAC	Replicase	489	Marais et al. 2016
	R2	CCATCAAGAACATGGTGCAGA			
ACLSV	F	TCTGCAAGAGAATTCAGTT	CP	800	Lu et al. 2015
	R	GTCTACAGGCTATTTATTATAAG			
CGRMV	F	AGAGGGCATTGGTTACTGGT	CP	988	This study
	R	AAACGGGCACTCTGGTAAGA			
CNRMV	F	CTTCCAAGCAAGATTGTCGC	CP	1172	This study
	R	TTCGCCCCGTGTTGTAAGAC			
GRGV	F1	CACCCCTTCTCCCCATGTGAC	CP	441	Fan et al. 2016
	R1	ACAGCTGGGGCTTCAGGGTT			
	F2	TCCGCCCCGAGAAGCACTTTG	RdRp	233	Fan et al. 2016
	R2	AGAAGCTCGGAGAGGAGAGGG			
PBNSPaV	F	AGACAATCGATGCCTGCAAC	CP	1046	This study
	R	AGCCCTTCTAACAGTCACGT			
PeVD	F	CCGACTCTGTGATTGACTACCC	Replicase Polyprotein	695	This study
	R	GAGGACTCGTGCGGACTTTA			
NSPaV	F1	CACTGTGATCCCATCCCCTT	RdRp	1036	This study
	R1	AAACCAGAGTCCGATCCGAG			
	F2	AACATGTGAGGTGGTGGAT	RdRp P1-P2 fusion	712	Lu et al. 2017
	R2	GCCCACATCTTCGACGATTC			
	F3	CCTGTACAGCCGAAGAAAGC	CP	474	Lu et al., 2017
	R3	ATGAGTGTGCAGGGTGATGA			

PaLV	F1	ATCCAGCATGATTCGTTCGT	RdRp	981	This study
	R1	CGTGACAGAGCCCATCTAAAA			
	F2	ACGATAACAATTCACCGCGC	CP	647	This study
	R2	GAGGAGCTGCGTCTACCTAG			
PLMVd	F	GGAATTCCTGTGATCCAGGTAC CGCCGTAGAAACT	Genome	337	Ambros et al. 1998
	R	CTGGATCACACCCCCCTCGGAA CCAACCGCT			

^a CP, Coat protein; RdRp, RNA-dependent RNA polymerase.

^bReferences:

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Table S3 Oligonucleotide primers used for amplification of NSPaV genomic sequences.

Primer		Primer sequence (5'-3')	Position	Product size (bp)
NSPaV-P1	F	CACTGTGATCCCATCCCCTT	108-127	1036
	R	AAACCAGAGTCCGATCCGAG	1143-1124	
NSPaV-P2	F	ATGACACGCTGGATGAC	529-545	2422
	R	GCTGTAGCTGCTTGCTG	2950-2934	
NSPaV-P3	F	AAGCGGCAAAGAAGAAATCC	2882-2901	1338
	R	TAGTCGCAACTGACCGTGGG	4219-4200	
NSPaV-P4	F	ATCTGGGAATTACAACGGACAT	3672-3693	1014
	R	CAACGTGGACCTTTGTTTGC	4685-4666	

Table S4 Infection by individual viruses and viroids was verified in 36 nectarine samples by RT-PCR.

Samples	Collecting Greenhouse	Cultivar	Tree age	Leaf symptoms	Fruit symptoms	RT-PCR results											
						APV1	APV2	APV3	ACLSV	CGRMV	CNRMV	GRGV	PBNSPaV	NSPaV	PaLV	PeVD	PLMVd
P1	#3	Chaoyue 1	2	Asymptomatic	Asymptomatic	-	-	-	-	-	-	-	-	-	-	-	+
P2	#3	Chaoyue 1	2	Asymptomatic	Asymptomatic	-	-	-	-	-	-	-	-	-	-	-	+
P3	#3	Chaoyue 1	2	Asymptomatic	Asymptomatic	-	-	-	-	-	-	-	-	-	-	-	+
P12	#2	Zhongyou 4	5	Chlorotic mottling	Asymptomatic	-	-	-	-	-	-	-	-	-	-	-	+
P13	#2	Zhongyou 4	5	Chlorotic mottling	Asymptomatic	-	-	-	-	-	-	-	-	-	-	-	+
P14(T05)	#2	Zhongyou 4	5	Chlorotic mottling	Asymptomatic	-	-	-	-	-	-	-	-	-	-	-	+
P15	#2	Zhongyou 4	5	Chlorotic mottling	Asymptomatic	-	-	-	-	-	-	-	+	-	-	-	+
P16	#2	Zhongyou 4	5	Asymptomatic	unknown	-	-	-	-	-	-	-	-	+	-	-	+
P17	#2	Zhongyou 4	5	Asymptomatic	unknown	-	-	-	-	-	-	-	-	+	-	-	+
P18	#2	Zhongyou 4	5	Asymptomatic	unknown	-	-	-	-	-	-	-	-	+	-	-	+
P19	#2	Zhongyou 4	5	Asymptomatic	unknown	-	-	-	-	-	-	-	-	+	-	-	+
P25(T03)	#2	Zhongyou 4	5	Asymptomatic	Asymptomatic	-	-	-	-	-	-	-	-	+	-	-	+
P25-F(S06)	#2	Zhongyou 4	5	Asymptomatic	Asymptomatic	-	-	-	-	-	-	-	-	+	-	-	+
P26	#2	Zhongyou 4	5	Asymptomatic	Asymptomatic	-	-	-	-	-	-	-	-	+	-	-	+
P26-F	#2	Zhongyou 4	5	Asymptomatic	Asymptomatic	-	-	-	-	-	-	-	-	+	-	-	+
P27	#2	Zhongyou 4	5	Asymptomatic	Asymptomatic	-	-	-	-	-	-	-	-	+	-	-	+
P20	#2	Zhongyou 4	5	Asymptomatic	dimpling	-	+	-	+	-	-	-	-	+	-	-	+
P20-F	#2	Zhongyou 4	5	Asymptomatic	dimpling	-	+	-	+	-	-	-	-	+	-	-	+
P21(T04)	#2	Zhongyou 4	5	Asymptomatic	dimpling	-	+	-	+	-	-	-	-	+	-	-	+
P21-F(S05)	#2	Zhongyou 4	5	Asymptomatic	dimpling	-	+	-	+	-	-	-	-	+	-	-	+

continue

Samples	Collecting Greenhouse	Cultivar	Tree age	Leaf symptoms	Fruit symptoms	RT-PCR results											
						APV1	APV2	APV3	ACLSV	CGRMV	CNRMV	GRGV	PBNSPaV	NSPaV	PaLV	PeVD	PLMVd
P23	#2	Zhongyou 4	5	Asymptomatic	dimpling	-	+	-	+	-	-	-	-	+	-	-	+
P23-F	#2	Zhongyou 4	5	Asymptomatic	dimpling	-	+	-	+	-	-	-	-	+	-	-	+
P24	#2	Zhongyou 4	5	Asymptomatic	dimpling	-	+	-	+	-	-	-	-	+	-	-	+
P24-F	#2	Zhongyou 4	5	Asymptomatic	dimpling	-	+	-	+	-	-	-	-	+	-	-	+
N8(T01)	#1	Youtao 1233	10	Bleached	Asymptomatic	+	-	-	+	-	-	-	+	+	+	+	+
N8-2	#1	Youtao 1233	10	Bleached	Asymptomatic	+	-	-	+	-	-	-	+	+	+	+	+
N9(T02)	#1	Youtao 1233	10	Bleached	pitting	+	-	-	+	+	+	-	+	+	+	+	+
N9-2	#1	Youtao 1233	10	Bleached	pitting	+	-	-	+	+	+	-	+	+	+	+	+
N7-2	#1	Youtao 126	10	Bleached	unknown	-	-	-	-	-	-	-	-	-	+	-	+
N7(S09)	#1	Youtao 126	10	Bleached	unknown	-	-	-	-	-	-	-	-	-	+	-	+
Y3-1	#1	Youtao 126	10	Bleached	unknown	-	-	-	-	-	-	-	-	-	-	-	+
Y3-2	#1	Youtao 126	10	Symptomless leaf of bleached	unknown	-	-	-	-	-	-	-	-	-	-	-	+
Y4	#1	Youtao 126	10	Bleached	unknown	-	-	-	-	-	-	-	-	-	+	-	+
Y6	#1	Youtao 126	10	Bleached	unknown	-	-	-	-	-	-	-	-	-	-	-	+
Y7	#1	Youtao 126	10	Bleached	unknown	+	+	-	+	-	-	-	-	-	+	+	+
Y8	#1	Youtao 126	10	Bleached	unknown	-	-	-	-	-	-	-	-	-	-	-	+

Of the 36 samples, P20-F, P21-F, P23-F, P24-F, P25-F, and P26-F were from fruit tissue. P14(T03), P25(T04), P21(T05), N8(T01), N9(T02), N7(S09), P25-F(S06), and P21-F(S05) were also screened for viruses and viroids by sequencing the small RNAs using next generation sequencing (NGS). ‘+’, positive. ‘-’, negative.