

Supplementary TABLE S1. Primers used for the detection and the molecular characterization of lettuce mottle virus (LeMoV) isolates JG2, P22 and SP, of lettuce star mosaic virus (LSMV) isolates JG1-A and JG1-B and of lettuce waikavirus 1 (LWV1) isolate JG1.

Targeted virus	Targeted gene	Primer name	5'-3' sequence	Genome positions	Annealing Temperature
<u>Detection primers</u>					
LWV1	RdRp	New-waika-F	GCTAGACATTGCTTGATTAATGC	10007-10029	58°C
		New-waika-R	CTGAACTTATAGGCACGTGCG	10391-10371	
LSMV-JG1A	RdRp	NSeco-varA-F	ATCTTTTTCCCAATTGGCTTC	7611-7632	58°C
		NSeco-varA-R	CACCCCAAAGCTTCCTTCAAG	8055-8034	
LSMV-JG1B	RdRp	NSeco-varB-F	AGAGAGTGGACGGAACCTGC	7294-7313	60°C
		NSeco-varB-R	ATCGAATCAAGAATGTTGCCG	7839-7818	
LeMoV	RdRp	LeMoV 3	ACATGAGCACTAGTGAGG	7225-7242	52°C
		LeMoV 4 multi	GCCTGCAAATAGAAGAGG	7891-7874	
<u>Characterization primers</u>					
LWV1	5' end	5Race-NWaika	CAGTAATAAGAGTGATCGATTCCCCATGCC	240-211	70°C
LWV1	3' end	3Race-NWaika	GGCGTTTGTAGCGCTGACGGGTG	11728-11750	60°C
LSMV-JG1B	5' end	5Race-NSecoB	CCTTACCCTTTGAGTCTTTGGTTGGCGC	496-469	56°C
LSMV-JG1B	3' end	3Race3-NSecoB	TCAAGTTGCGATTCTTTCGTTTCAGACG	9133-9160	60°C
LeMoV-JG2	5' end	UPM-GTTTAAAG	AACGCAGAGTACATGGGGTTTAAAG	1-8	65°C
		LeMoV-5Race1	CCCTGCAGCGCTCCACTGAGAGGACC	414-389	65°C
		LeMoV-5Race2	AAGCACTGAAAGCAAACCTCCTCGATAGC	261-234	65°C
LeMoV-JG2	3' end	LeMoV-3Race1	CTGCTTGGCGACGACTTGACTTTACCG	9084-9110	62°C
		LeMoV-3Race3	GCGGTAGCCAATCCTCTTGAATGAGGG	9630-9656	70°C
		LeMoV-gap-R1	CAATTAAGGTCTTCCGTACGC	9282-9262	62°C
		LeMoV-gap-R2	CCCTCATTCAAGAGGATTGGCTACCGC	9656-9630	70°C
LeMoV-P22	CP1	P22secoF1	GTGGACACAGTTGAGTGGAAAT	1194-1215	55°C
		P22secoR1	TTCATGTGGTCAATGCAGAC	1403-1383	
LeMoV-P22	CP2	P22secoF2	GCCAGCAGAAATCAGAAGCATA	1781-1802	55°C
		P22secoR2	CTGGAAATGGCTGTTCTATATC	2125-2103	
LeMoV-P22	NTP Domain	P22secoF3	CGCTACACACACTCGGCTCCACC	3126-3148	58°C
		P22secoR3	CAAGGATCAGACTGGTGATTGGT	3432-3410	
LeMoV-P22	NTP Domain	P22secoF4	GAGTACCGTGTTGATGAACCT	3572-3592	55°C
		P22secoR4	ATTGCCATAATAACTCTGTAGA	3962-3940	

LeMoV-P22	NTP Domain	P22secoF5	CGCTTCGAGGGAAGTCGTTGG	4846-4866	55°C
		P22secoR5	CTTCCCTTGTCACTTCCATTC	5016-4996	
LeMoV-P22	NTP Domain	P22secoR6	ACACGTCGCTGAAGATGGAG	3369-3350	58°C
		P22secoF3	CGCTACACACACTCGGCTCCACC	3126-3148	
LeMoV-P22	RdRp	P22secoF7	AGCGCCGAAGACCCTCGTTTG	6807-6827	65°C
		P22secoR7	TCGTACTCATATTGATGCTC	7059-7040	
LeMoV-P22a	P1	LmoA-F	ATGACGTTAGCAATCCAACC	1-20	52°C
		LmoB-R	GCCGTTTCGAGCCATCC	787-771	
LeMoV-P22a	CP1	LmoE-F	GCAATGAAGCTTTCAGTGGC	1354-1373	55°C
		LmoF-R	CCTCTGTCCCAAGTGATTC	2182-2163	
LeMoV-P22a	CP2-NTB	LmoG-F	CAGCATCACTCCCAAAGG	2030-2047	53°C
		LmoJ-R	GACTGGTGATTGGTGTTGC	3423-3405	
LeMoV-P22a	Protease-RdRp	LmoS-F	ACGGCGCGCATTGAGAGG	5832-5849	53°C
		LmoV-R	GTTTCTCCTCGAGCCCCAG	6621-6603	
LeMoV-P22a	RdRp	LmoW-F	ATGGAAAGAAGTGGCTG	7102-7118	52°C
		LmoX-R	GAARAGAGGAGACTGTGC	7875-7858	
LeMoV-P22a	RdRp	LmoY-F	AGCGTGGATCCAGTTGGC	7715-7732	55°C
		LmoZ-R	GATACCACCAAGAGAGTCC	8405-8387	
LeMoV-SP	P1-CP1	P6-F	GCTGCTCAGGAAAGACAACG	620-639	55°C
		P10-R	ACTCTGCTCAGGGCGTTG	1570-1553	
LeMoV-SP	NTB	P11-F	CTCATGATGATCAGCGC	2767-2783	55°C
LeMoV-P22a		M1-R	GGTACTCAGGGCTGTTGG	3771-3754	
LeMoV-P22a	NTB-Protease	M7-F	GCTTGTTGGACTTGCC	5252-5267	53°C
LeMoV-SP		P16-R	CATGTCGTCCTCTCAGC	6047-6030	
LeMoV-SP	NTB	P18-F	GTGTCGCTTTGAGTGTC	3535-3551	52°C
		P20-R	CATTGCCGCAGTCGTCAT	5372-5355	
LeMoV-P22a	NTB	M12-R	GTCCATGCTTGCTTCCAC	4807-4790	52°C
		P18-F	GTGTCGCTTTGAGTGTC	3535-3551	

RdRp: RNA dependent RNA polymerase; CP1: coat protein 1; CP2: coat protein 2; P1: protein P1; NTB : nucleotide triphosphate binding motif of the viral helicase.

Supplementary TABLE S2. Pairwise percentages of amino acid identity for the CP (lower diagonal) and Pro-Pol (upper diagonal) regions of the polyproteins of *Waikavirus* genus members.

	1	2	3	4	5	6	7
1 BVCV NC_027915	-	63	56	69	68	41	64
2 RTSV NC_001632	45	-	58	62	62	43	60
3 MCDV NC_003626	41	38	-	60	57	40	52
4 RCaV1 MH325329	51	44	39	-	65	40	61
5 BnV1 NC_040586	61	47	43	53	-	42	64
6 PWV LC488189	25	25	25	23	25	-	41
7 LWV1-JG1 MT348710	56	44	42	53	55	24	-

Supplementary TABLE S3. Pairwise percentages of identity for the CP (lower diagonal) and Prot-Pol (upper diagonal) regions of the polyproteins of *Sequivirus* genus members.

	1	2	3	4	5	6	7	8	9
1 PYFV NC_003628	-	76	60	58	59	61	62	59	60
2 CNDV NC_038320	75	-	61	61	61	61	60	59	60
3 LeMoV-P22 MT348708	52	53	-	97	96	76	76	73	72
4 LeMoV-JG2 MT348707	52	53	100	-	96	76	76	71	71
5 LeMoV-SP MT348709	52	53	98	98	-	75	76	72	71
6 LSMV-JG1-A MT348705	51	53	83	83	83	-	91	73	73
7 LSMV-JG1-B MT348706	51	53	83	83	83	98	-	74	73
8 DaYMV-RNA24P-1 MT559283	52	51	72	72	72	73	74	-	88
9 DaYMV-RNA24P-2 MT559284	51	52	72	72	72	73	73	94	-

Supplementary TABLE S4. Description of the viral isolates used for the phylogenetic studies

Virus	Genus	Acronym	Accession number
Carrot necrotic dieback virus	<i>Sequivirus</i>	CNDV	NC_038320
Dandelion yellow mosaic virus	<i>Sequivirus</i>	DaYMV	DQ675189
Lettuce mottle virus	<i>Sequivirus</i>	LeMoV	DQ675190, DQ675191, AM039965
Parsnip yellow fleck virus	<i>Sequivirus</i>	PYFV	NC_003628
Bellflower vein chlorosis virus	<i>Waikavirus</i>	BVCV	NC_027915
Brassica napus virus 1	<i>Waikavirus</i>	BnV1	NC_040586
Maize chlorotic dwarf virus	<i>Waikavirus</i>	MCDV	NC_003626
Persimmon waikavirus	<i>Waikavirus</i>	PWV	LC488189
Red clover associated virus 1	<i>Waikavirus</i>	RCaV1	MH325329
Rice tungrol sperical virus	<i>Waikavirus</i>	RTSV	NC_001632
Grapevine fanleaf virus	<i>Nepovirus</i> , subgroup A	GFLV	NC_003615, NC_003623
Tomato black ring virus	<i>Nepovirus</i> , subgroup B	TBRV	NC_004439, NC_004440
Blackcurrant reversion virus	<i>Nepovirus</i> , subgroup C	BRV	NC_003509, NC_003502
Cowpea mosaic virus	<i>Comovirus</i>	CPMV	NC_03549, NC_003550
Broad bean wilt virus 1	<i>Fabavirus</i>	BBWV1	NC_05289, NC_005290
Cherry leaf roll virus	<i>Cheravirus</i>	CLRV	NC_06271, NC_006272
Strawberry latent ringspot virus	Unassigned	SLRSV	NC_06964, NC_006965
Carrot torradovirus 1	<i>Torradovirus</i>	CaTV1	NC_25479, NC_025480
Strawberry mottle virus	<i>Stramovirus</i>	SMoV	NC_03445, NC_03446
Satsuma dwarf virus	<i>Satsumavirus</i>	SDV	NC_03785, NC_03786
Chocolate lily virus A	<i>Cholivirus</i>	CLVA	NC_16443, NC_16444

Supplementary Figure 1. Multiple alignment of the predicted short overlapping ORFX from all members of the genus *Waikavirus*.

BVCV NC_027915	MQR-VLLIISLAVNMIALFLFTLGLILKLPVILIVGVFVIMLNIFLSVLALVIKPEEN-----GSQFLERVITGTPLAR---VAVPRAPLPRPVRV*
MCDV NC_003626	MQR-VLLIIGLAVNMLSCLFFQMLGLVFRPLLLVIGLCVIMLNIFLSVLALVTRSEDT-----LLDLMTRMHIQTPTAQNVA PVARETLGPRSR*--
RCaV1 MH325329	MQR-ALLVVG LAVNMIALFLFTLGLILRIPVILVVGIFVIMLNIFLSVLALVTKPEED-----ISQFIERITSGTPLAR---TVRRPAPSRAPERG*
PWV LC488189	MLK-IAIISGLVSLVMAICLICMGLMHTPAILLVGVLCFAALMLLVVGVLVVQDDVGQNTFSMGIPEWLS-PRRAPPVVPG-ARPA PRVARPSGMRA*
BnV1 NC_040586	MQR-VLLIASLAINMAALFMQVLG LLLKQPIIILIVGICVIMLNIFLNVLALVTKPEED-----FSQFLERASAGTPLAR---NAERRAPLPVRERR*
RTSV NC_001632	MQRGFYLIICLSLSMIALFVLM LALVFRKPVLMITLCVIMLSIFLNVLALVVRPEEN-----FSELVARVSTGTPLAR---LAERRVDLPTRGRI*
LWV1-JG1 MT348710	MQR-VLLIISLGVNMIALFLGCMGLILKNPIIMIVGVFVIMLNIFLNVLALVIKPEEN-----ISQFMDRVSAGTPLAR---RAERPAPLPARGRA*

BVCV: bellflower vein chlorosis virus; MCDV: maize chlorotic dwarf virus; RcaV1: red clover associated virus 1; PWV: persimmon waikavirus; BnV1: Brassica napus virus 1; RTSV: rice tungrol sperical virus; LWV1: lettuce waikavirus 1.