```
FBNYV-[ES;R4]_DNA-U2 KC979033        435-GCTAACGAAGAAGCGTTAGGGGAGTTTTGTAGATATCACGGAAGGAGAGTAC-486
FBNYV-[ES;Mu29D]_DNA-U2 KC979025     435-GCTAACGAAGAAGCGTTAGGGGAGTTTTGTAGATATCACGGAAGGAGAGTAC-486
FBNYV-[ES;R7]_DNA-U2 KC979041        435-GCTAACGAAGAAGCGTTAGGGGAGTTTCGTAGATATCACGGAAGGAGAGTAC-486
FBNYV-[AZ;13.5]_DNA-U2 KC979015      435-GCTAACGAAGAAGCGTTAGGGGAGTTTTGTAGATACCATGGAAGGAGAGTAG-486
FBNYV-[MA;23]_DNA-U2 GQ274029        435-GCTAACGAAGAAGCCTTAGGAGAGTTTTGTAGATATCATGGAAGGATAGTAT-486
FBNYV-[SY;292-88]_DNA-U2 Y11409      435-GCTAACGAAGAAGCGTTAGGAGAGTTTTGTAGATATCATGGAAGGATAGTAT-486
FBNYV-[EG;1-93]_DNA-U2 AJ132184      435-GCTAACGAAGAAGCGTTAGGAGAGTTTTGTAGATACCATGGAAGGAGAATAT-486
FBNYV-[AZ;12]_DNA-U2 KC979006        435-GCTAACGAAGAAGCGTTAGGAGAGTTTTGTAGATATCATGGAAGGAGAGTAC-486
FBNSV-[AZ;1]_DNA-U2 KC978972         431-GCCCATGAAGATGTTCTTGGTGAATTCTGCAGACGCCATGGAAGAAGAGTTC-482
FBNSV-[AZ;10]_DNA-U2 KC978980        431-GCCCATGAAGATGTTCTTGGTGAATTCTGCAGACGCCATGGAAGAAGAGTTC-482
FBNSV-[MA;5]_DNA-U2 GQ274037         425-GCATACGAAGATGTTCTTGGAGAATTTTGCAGACGCCATGGAAGAAGAGTCC-476
FBNSV-[ET;Hol-97]_DNA-U2 GU983872    429-GCCCACGAAGATGTTCTTGGTGAATTCTGCAGACTCCATGGAAGAAGAGTTC-480
FBNSV-[AZ;15]_DNA-U2 KC978998        430-GCCCATGAAGATGTTCTTGGTGAATTCTGCAGACGCCATGGAAGAAGAGTTC-481
FBYLV-[ET;231]_DNA-U2 HE654129       445-GCTCACGAAGATGTTCTTGGGGAAATATGCAGATATCATGGAAGAAGAGTTA-496
MDV-[JP;1]_DNA-U2 AB000926           422-CGCAATGAAGATGTTCTTGGAGAGATGTGCAGACGCCATGGAAGGAAATTGC-473
BMLRV-[AT;3]_DNA-U2 KC978945         452-TCCAATGAAGATATGTTGGGTCAGTTCTGCAGACGCCATGGAAGAAGAGTCA-503
BMLRV-[SE;153]_DNA-U2 KC978964       422-TCCAATGAAGATATGCTTGGTCAGTTCTGCAGACGCCATGGAAGAAGGCTCA-473
BMLRV-[AZ;47]_DNA-U2 KC978955        425-TCTAACGAAGATATGTTGGGTCAGTTCTGCAGACGCCATGGAAGAAGGCTCA-476
PYSV-[AT;15]_DNA-U2 KC979059         500-TCAAATGAAGATATGCTTGGCAAGTTCTGCAGACGCCATGGAAGAAGCGTCA-451
PNYDV-[DE;15]_DNA-U2 JN133284        378-TTTAATGAAGATATGGTTGGAATTCGCTGTCGTCGTCATGGAAGAAGAGTGA-440
PNYDV-[AT;1]_DNA-U2 KC979049         379-TTTAATGAAGATATGGTTGGAATTCGCTGTCGTCGTCATGGAAGAAGAGTGA-441
nanoU2dir primer                                                  TGYAGAYRYCATGGAAGRA
```

Figure S1. Multialignment of nanovirus DNA-U2 sequences. Alignment of multiple sequences was performed with MultAlign (INRA, Toulouse, URL: http://multalin.toulouse.inra.fr/multalin/). Sequences displayed span nucleotides from the respective 5'- to 3'-coordinates indicated. The sequence of the degenerate primer nanoU2dir is shown below the alignment

Figure S2. Multialignment of nanovirus DNA-U4 sequences. Alignment of multiple sequences was performed with MultAlign (INRA, Toulouse, URL: http://multalin.toulouse.inra.fr/multalin/). Sequences displayed span nucleotides from the respective 5'- to 3'-coordinates indicated. The sequence of the degenerate primer nanoU4dir is shown below the alignment.

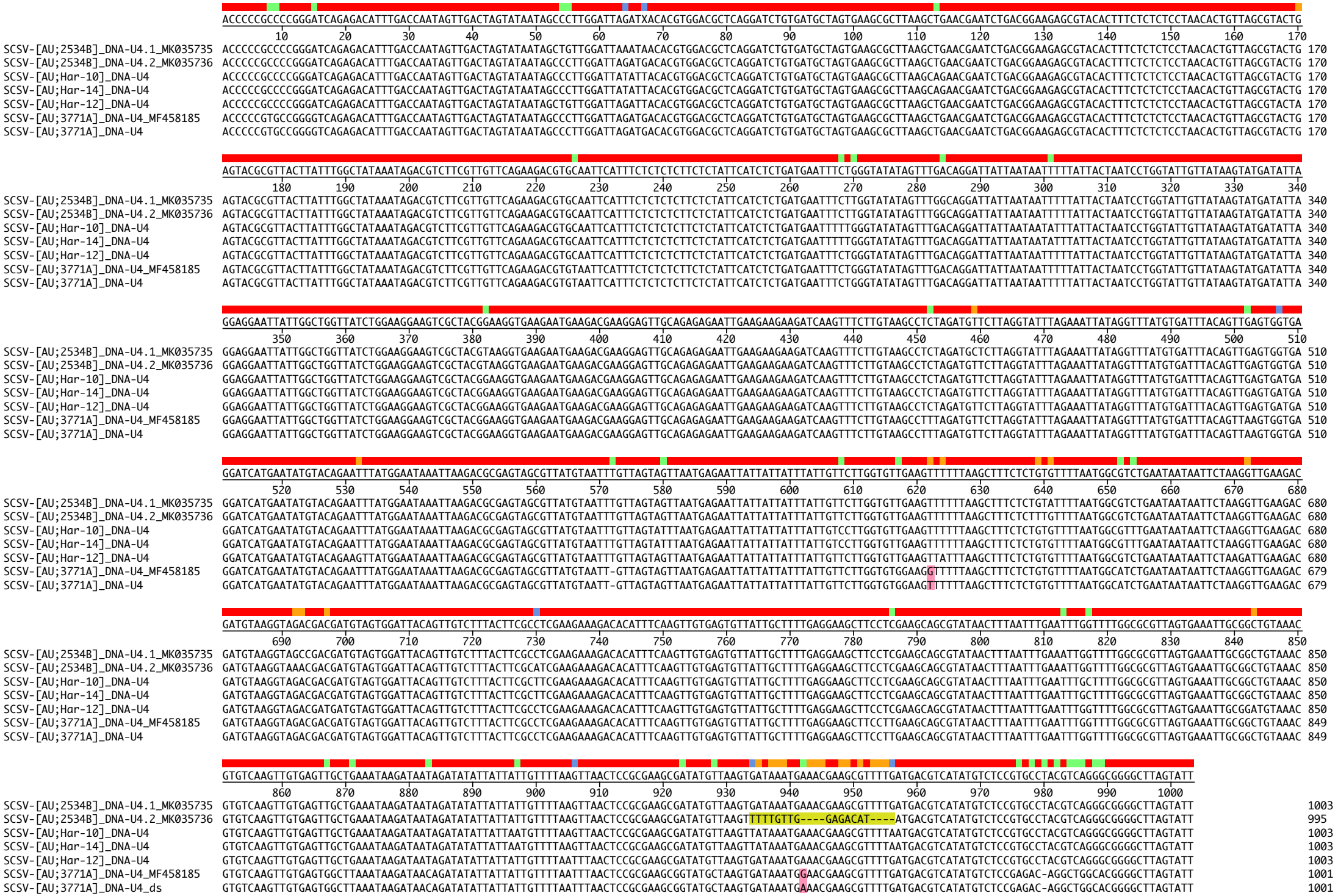

Figure S3. Multialignment of SCSV DNA-U4 sequences. Alignment was performed using MegAlign of DNA Star. The yellow box indicates sequence homologous to DNA-C. Pink boxes indicate nucléotide differences between two DNA-U4 sequences from SCSV-[AU;3771A], that of a cloned DNA (DNA-U4_MF458185) and that of the DNA-U4 consensus sequence determined by deep sequencing (DNA-U4_ds).

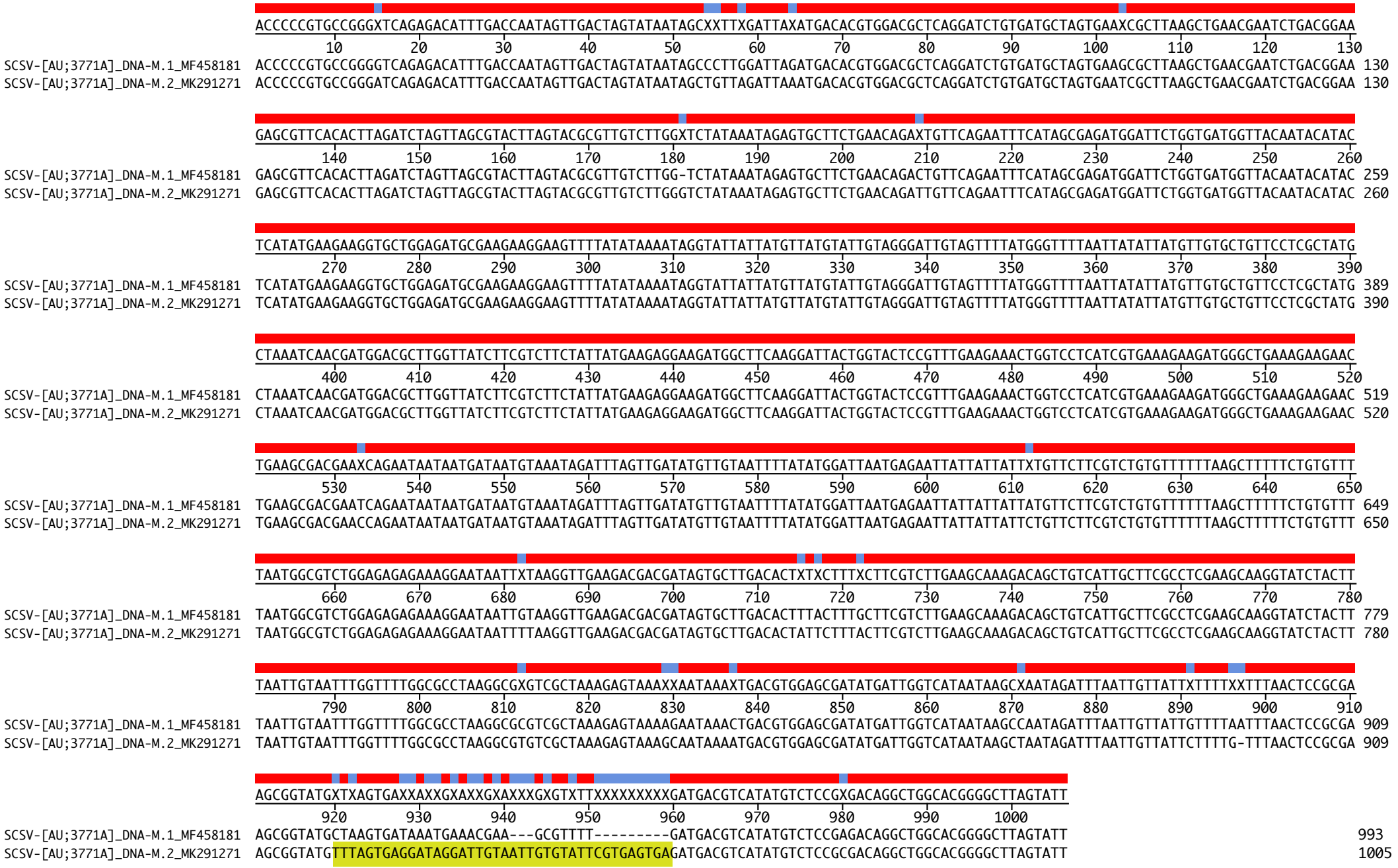

Figure S4. SCSV-[AU;3771A]_DNA-M.1_MF458181_vs_DNA-M.2_MK291271 alignment. Alignment was performed using MegAlign of DNA Star. The yellow box indicates sequence homologous to DNA-R.

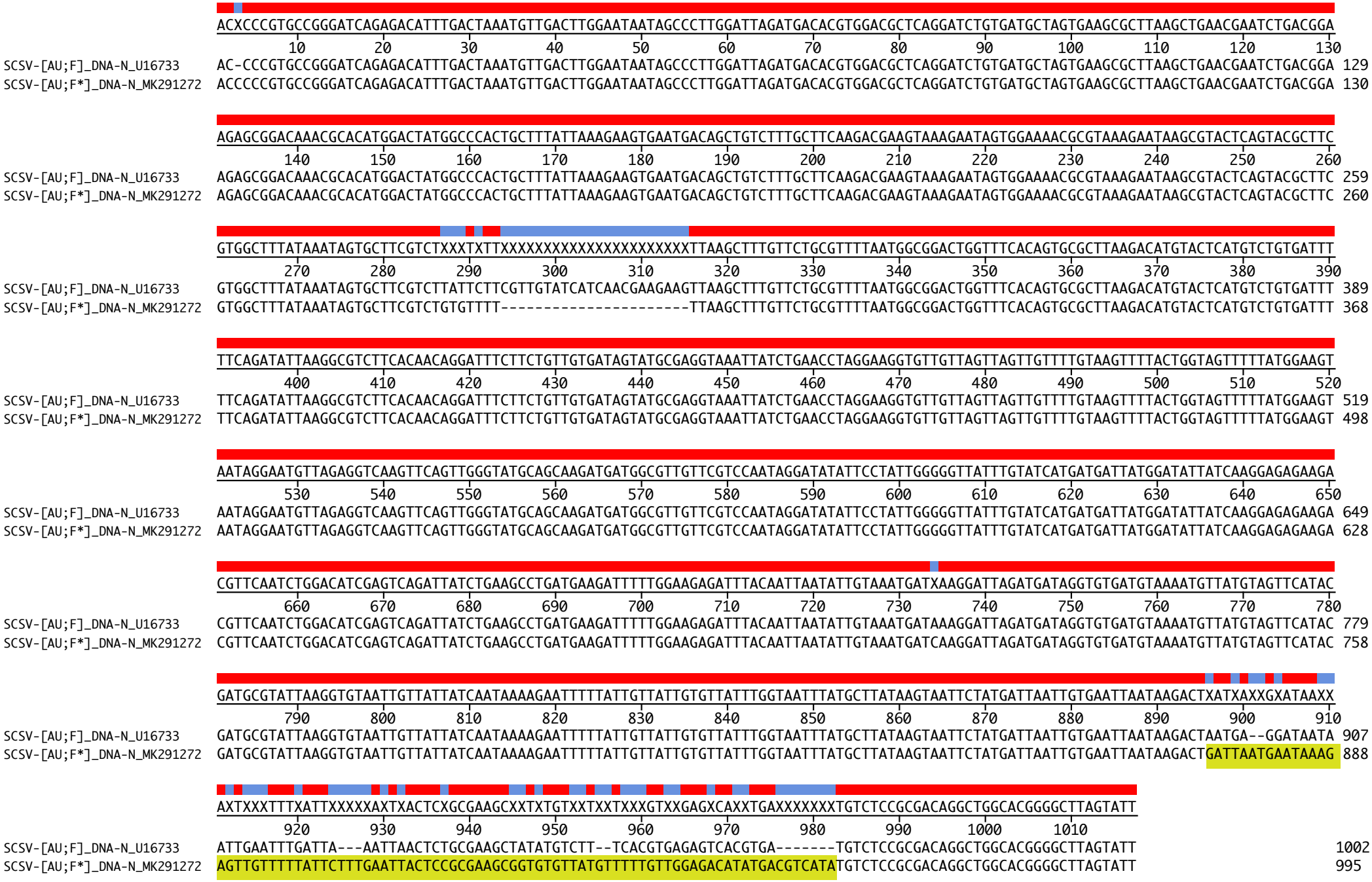

Figure S5. SCSV-[AU;F]_DNA-N_U16733 vs SCSV-[AU;F*]_DNA-N_MK291272 alignment. Alignment was performed using MegAlign of DNA Star. The yellow box indicates sequence homologous to DNA-S.

Table S1. Primers used for amplification of SCSV DNAs

DNA	Primer name (direction)	Sequence (5'-3')	Tm, C°	Use [a, b]
nanovirus DNAs [c]	nano-STL-dir	GGCTTAGATATTACCCC	49.2	2
	nano-STL-rev	GGTAATACTAAGCCCC	49.2	2 [f]
nanovirus DNA-U2 [d]	nano-U2-dir	TGYAGAYRYCATGGAAGRA	53.4	2 [f]
nanovirus DNA-U4 [e]	nano-U4-dir	GGTTGAHGRHGAHGAYSTGGARTA	61.8	2 [f]
R	SCSV-R-dir	TTCGTCTACAGTCCTTCGTCTCCATCAA	65.1	1, 4
	SCSV-R-rev	GTAGACGAATAATTTGGGTGTATGGCCC	65.1	1, 4
	SCSV-R790-dir	GAATACGTCAATTATGGAGTCATTGAACAGCT	64.4	1
	SCSV-R790-rev	ACGTATTCCTCGAAGCTTCGAGGAAAGTCG	65.5	1
M	SCSV-M-dir	GAAGATGGGCTGAAAGAAGAACTGAAGCG	66.7	1, 4
	SCSV-M-rev	CCATCTTCTTTCACGATGAGGACCAGTTTC	66.8	1, 4
	SCSV-M390-dir	CTAAATCAACGATGGACGCTTGGTTATC	63.7	1
	SCSV-M390-rev	TGATTTAGCATAGCGAGGAACAGCACAAC	65.3	1
N	SCSV-N-dir	GGAAGGTGTTGTTAGTTAGTTGTTTTGTAA	61.3	1, 4
	SCSV-N-rev	CACCTTCCTAGGTTCAGATAATTTACCTCG	65.4	1, 4
	SCSV-N420-dir	GCTTCGTGGCTCTATAAATAGTGCTTCGTCTT	63.4	1
	SCSV-N420-rev	CACGAAGCGTACTGAGTACGCTTATTCTTTACG	65.0	1
C	SCSV-C-dir	GAGTTATGGAGGAGATGCATTACAAGCTT	63.9	1, 4
	SCSV-C-rev	CCATAACTCTCCATTTCAGATTTA	55.9	1, 4
S	SCSV-S-dir	GGAGGTTAAGCCATTCGCTGATGGTTCAAG	68.1	1, 4
	SCSV-S-rev	TAACCTCCGCTCCAGTAACATCTCTCTTAT	65.4	1, 4
U1	SCSV-U1-dir	GTGATTAACGTGAAGGTACTGAGGTT	61.6	1, 4
	SCSV-U1-rev	CGTTAATCACTTGAACCTTCTC	56.5	1, 4
U2	SCSV_37-U2frDir	CTAAGATGCCTAGAGATGGACCTGCAAGGTG	69.5	1
	SCSV_37-U2frRev	CTTAGGATAAGCCTTCACTCTTCTTCCATG	65.4	1
	SCSV_37-U2fr2dir	AATTATAGATATCAAGTGTATTACTCTG	56.3	1, 4
	SCSV_37-U2fr2rev	CTATAATTACCATAATCATACAAATCA	54.3	1, 4
	SCSV-U2-224-dir	GATTTGTTCTGGGTTTTATATGAA	55.3	1
	SCSV-U2-224-rev	CAGAACAAATCTTGTTCTTCCTT	54.2	1
U4	SCSV_37-U4fr2dir	GTGAGGATCATGAATATGTACAG	57.1	1, 4
	SCSV_37-U4fr2rev	TCCTCACCACTTAACTGTAAATCAC	59.7	1, 4
	SCSV-U4-340-dir	GGAGGAATTATTGGCTGGTTATCTGGAA	63.7	1,3, 4
	SCSV-U4-340-rev	ATTCCTCCTAATATCATACTTATAACAATACCAGG	63.6	1, 4
	SCSV-U4.1-932-rev	CGTTTCATTTATCACTTAACATATCGC	58.9	3
	SCSV-U4.2-932-rev	TCTCCAACAAAAACTTAACATATCGC	58.5	3

a – Primers used for PCR amplification of the corresponding DNAs with the purpose of: (1) entire component amplification and cloning, (2) partial amplification, (3) specific amplification of DNA-U4.1 or DNA-U4.2 fragment by PCR, (4) detection of SCSV DNAs in field samples of symptomatic subterranean clover.

b – PCR amplifications employing component-specific primers were performed at annealing temperatures corresponding to a minimal Tm of a given primer pair.

c – Primers designed based on the DNA sequence in the region with potential stem-loop structure (STL) of nanovirus DNAs

d – Primers designed on the basis of the alignment of the nanovirusDNA-U2 sequences.

e - Primers designed on the basis of the alignment of the nanovirus DNA-U4 sequences.

f - PCR amplifications using nano-STL-rev in combination with a degenerate primer were performed with Taq II DNA polymerase as follows: 95^{O} C, 5 min; 5 cycles of: 95^{O} C, 30 s, 45^{O} C, 45 s, 72^{O} C, 2 min; followed by 35 cycles of: 95^{O} C, 30 s, gradient 45^{O} C to 65^{O} C, 45 s, 72^{O} C, 45 s; final elongation: 72^{O} C, 4 min.

Table S2. Summary of deep sequencing data from three SCSV samples

	Sample / DNA	SCSV-[AU;3771A] (1,519,544 total reads) Reads	% of reads	Cov. Mean	SCSV-[AU; 2534B] (1,560,076 total reads) Reads	% of reads	Cov. Mean	SCSV-[AU;-F*] (1,984,716 total reads) Reads	% of reads	Cov. Mean
Genomic DNAs	DNA-R	4,273	0.28	6,878	32,450	2.08	6,156	26,548	1.34	4,881
	DNA-S	10,419	0.69	1,849	73,136	4.69	14,378	33,825	1.70	6,496
	DNA-M	27,366	1.80	4,394	68,165	4.37	12,132	400,292	20.17	72,184
	DNA-M.2 [a]	12,979	0.85	2,074						
	DNA-N	11,943	0.79	2,072	100,037	6.41	19,303	12,764	0.64	2,343
	DNA-C	20,471	1.35	3,602	43,620	2.80	8,152	6,168	0.31	796
	DNA-U1	5,727	0.38	929	40,129	2.57	7,436	121,430	6.12	22,534
	DNA-U2	2,510	0.17	421	41,474	2.66	7,863	2,513	0.13	431
	DNA-U4	86,676	5.70	14,167	109,090	6.99	19,932			
	DNA-U4.2				123,375	7.91	22,640			
alphasatellites	SCSA 1	169,370	11.15	29,851	293,801	18.83	57,449			
	SCSA 1.1							91,250	4.60	17,207
	SCSA 1.2							93,260	4.70	17,178
	SCSA 2	125,028	8.23	22,230	192,094	12.31	38,112	460,008	23.18	89,133
	SYSA 3	31,974	2.10	5,520						
	total	508,736	33.48		1,117,371	71.62		1,248,058	62.88	

Circularized consensus sequences were generated from *de novo* assembly of deep sequencing reads.
Cov. Mean – average coverage.
a – recombinant with DNA-R

Table S3. Nucleotide substitutions

Comparison of two different SCSV isolates	Total changes	% divergence	C/A	G/A	T/A	G/C	T/C	T/G	indel	other
SCSV-[AU;3771A]_DNA-R_MF458178 SCSV-[AU;2534B]_DNA-R_MK035728	12	1,2	1	3	1	1	4	2	0	
SCSV-[AU;3771A]_DNA-S_MF458179 SCSV-[AU;2534B]_DNA-S_MK035729	38	3,8	1	8	8	4	11	5	1	
SCSV-[AU;3771A]_DNA-C_MF458180 SCSV-[AU;2534B]_DNA-C_MK035730	21	1,7	0	5	4	2	3	3	4	
SCSV-[AU;3771A]_DNA-M_MF458181 SCSV-[AU;2534B]_DNA-M_MK035731	50	3,7	2	6	8	6	10	4	14	
SCSV-[AU;3771A_DNA-N_MF458182 SCSV-[AU;2534B]_DNA-N_MK035732	11	1	1	4	1	0	2	2	1	
SCSV-[AU;3771A]_DNA-U1_MF458183 SCSV-[AU;2534B]_DNA-U1_MK035733	26	2,5	2	9	2	4	5	2	2	
SCSV-AU;3771A]_DNA-U2_MF458184 SCSV-[AU;2534B]_DNA-U2_MK035734	15	1,4	1	4	1	2	3	3	1	
SCSV-[AU;3771A]_DNA-U4_MF458185 SCSV-[AU;2534B]_DNA-U4_MK035735	38	3,7	2	11	2	4	10	7	2	
Sum	**211**		**10**	**50**	**27**	**23**	**48**	**28**	**25**	
Comparison of cloned DNAs vs deep sequenced DNAs (_ds) of the same isolate (SCSV-[3771A])	**Total changes**	**% divergence**	**C/A**	**G/A**	**T/A**	**G/C**	**T/C**	**T/G**	**indel**	**other**
SCSV-[AU;3771A]_DNA-R_MF458178 SCSV-[AU;3771A]_DNA-R_ds	0	0	0	0	0	0	0	0	0	
SCSV-[AU;3771A]_DNA-S_MF458179 SCSV-[AU;3771A]_DNA-S_ds	5	0,5	0	0	1	0	3	1	0	
SCSV-[AU;3771A]_DNA-C_MF458180 SCSV-[AU;3771A]_DNA-C_ds	7	0,7	0	1	0	1	3	2	0	
SCSV-[AU;3771A]_DNA-M_MF458181 SCSV-[AU;3771A]_DNA-M_ds	2	0,2	0	0	0	0	0	2	0	
SCSV-[AU;3771A]_DNA-M_MF458181 SCSV-[AU;3771A]_DNA-M.2_ds (recombinant with DNA-R)	25	3,7	2	6	3	4	5	2	2 + 12†	† part of 32 nt derived from DNA-R
SCSV-[AU;3771A_DNA-N_MF458182 SCSV-[AU;3771A_DNA-N_ds	8	0,8	1	3	1	0	2	1	0	
SCSV-[AU;3771A]_DNA-U1_MF458183 SCSV-[AU;3771A]_DNA-U1_ds	15	1,4	1	5	1	3	3	1	1	
SCSV-AU;3771A]_DNA-U2_MF458184 SCSV-AU;3771A]_DNA-U2_ds	0	0	0	0	0	0	0	0	0	
SCSV-[AU;3771A]_DNA-U4_MF458185 SCSV-[AU;3771A]_DNA-U4_ds	2	0,2	0	1	0	0	0	1	0	
Sum, counting recombination event as 1	**64**		**4**	**16**	**6**	**8**	**16**	**10**	**1**	
Comparison of published SCSV-[AU;F] DNAs with deep sequenced SCSV-[AU;F*] progeny	**Total changes**	**% divergence**	**C/A**	**G/A**	**T/A**	**G/C**	**T/C**	**T/G**	**indel**	**other**
SCSV-[AU;F]_DNA-R_AJ290434 SCSV-[AU;F*]_DNA-R	0	0	0	0	0	0	0	0	0	
SCSV-[AU;F]_DNA-S_U16734 SCSV-[AU;F*]_DNA-S	12	1	1	1	2	1	2	3	2	
SCSV-[AU;F]_DNA-C_U16732 SCSV-[AU;F*]_DNA-C	3	0,3	0	0	0	2	1	0	0	
SCSV-[AU;F]_DNA-M_U16730 SCSV-[AU;F*]_DNA-M	29	1,8	2	4	1	2	6	3	11	
SCSV-[AU;F]_DNA-N_U16733 SCSV-[AU;F*]_DNA-N (Δ 22nt & 87nt recombinant with DNA-S)	36	3,6	2	9	8	0	6	8	1 + Δ22	87 nt recombinant with DNA-S
SCSV-[AU;F]_DNA-U1_U16736 SCSV-[AU;F*]_DNA-U1	11	0,8	1	3	1	0	3	0	3	
SCSV-[AU;F*]_DNA-U2_MF458186 (cloned DNA) SCSV-[AU;F*]_DNA-U2	3	0,3	2	1	0	0	0	0	0	
SCSA1-[AU;F]_alphasat_1_U16731 SCSA1-[AU;F*]_alphasat_1.1	3	0,3	0	0	1	0	1	1	0	
SCSA1-[AU;F]_alphasat_1_U16731 SCSA1-[AU;F*]_alphasat_1.2 (Δ24 nt)	4	0,3	1	0	1	0	1	0	Δ 24	
SCSA2-[AU;F]_alphasat_2_U16735 SCSA2-[AU;F*]_alphasat_2	1	0,1	1	0	0	0	0	0	0	
Sum, counting deletions >20 nt and recobination event as 1	**102**		**10**	**18**	**14**	**5**	**20**	**15**	**16**	

Table S5. Detection by PCR of SCSV DNAs in field samples of symptomatic subterranean clover

DNA / Sample	-R	-S		-M		-C		-N	-U1		-U2		-U4[a]		-U4[b]	
	x35[c]	x35	x43[d]	x35	x43	x35	x43	x43	x35	x43	x35	x43	x35	x43	x35	x43
1	-	-	nt[e]	-	nt	-	nt	-	-	nt	-	-	-	-	-	-
2	-	-	nt	-	nt	-	nt	-	-	nt	-	-	-	-	-	-
3	++	+	++	++	nt	-	++	+	+	nt	+	+	-/+	++	-	-/+
4	++	-	+	-	+	-	++	++	-	++	-	+	-	-	-	-
5	++	-	++	-	+	-	-/+	-	-	-/+	-	+/-	-	-	-	-
6	+++	+++	nt	+++	nt	+++	nt	+++	+++	nt	+++	+++	+++	+++	+	++
7	++	+	nt	++	nt	++	nt	+	+/-	nt	+	+	-/+	+	-/+	-/+
8	-	-	nt	-	nt	-	nt	-	-	nt	-	-	-	-	-	-
9	-	-	nt	-	nt	-	nt	-	-	nt	-	-	-	-	-	-
10	++	-	++	++	nt	-	++	+	-	++	+	++	-	+	+	-/+
11	++	+	nt	++	nt	-	-	+	+	nt	+	++	-/+	++	-/+	+
12	++	++	nt	++	nt	++	nt	++	++	nt	+	++	+	+	+	+
13	++	-	-	-	++	-	-	-	-	+/-	-	+	-	+/-	-	-
14	++	++	nt	++	nt	++	nt	++	++	nt	++	++		++	+	+
Total SCSV-positive samples	10	9		10		8		8	10		10		8		7	

Subterranean clover plants showing symptoms of potential SCSV infection were collected on December 24, 2018, in Harrison, ACT. After total DNA extractions, PCR reactions were performed using primers specific for SCSV DNA (indicated in Table S1), the amplified DNAs were subjected to agarose gel electrophoresis. The relative amounts of amplification products from different samples were estimated visually by staining intensity, ranging from traces (-/+) to very high amplification levels (+++).
a – primers SCSV_37-U4fr2dir and SCSV_37-U4fr2rev.
b – primers SCSV-U4-340-dir and SCSV-U4-340-rev.
c - 35 cycles of PCR amplification.
d - 43 cycles of PCR amplification.
e - if 35 PCR cycles yielded amplified DNA of a given genome component, no amplification using 43 cycles was done.

Table S6. Detection of SCSV DNAs in field samples after rolling circle amplification followed by PCR.

DNA / Sample	-S	-C	-N	-U4[b]
1	+++	+++	+++	+++
4	+++	+++	+++	+++
5	+++	+++	+++	+++
11	+++	+++	+++	+++
13	+++	+++	+++	+++

Total DNA extractions of samples 1, 4, 5, 11 and 13 (same as in Table S5) were subjected to rolling circle amplification followed by PCR amplification (35 cycles) using the same SCSV component-specific primers as in Table S5.