

Supplementary materials

Cultivated tomato (*Solanum lycopersicum* L.) suffered a severe cytoplasmic bottleneck during domestication: implications from chloroplast genomes

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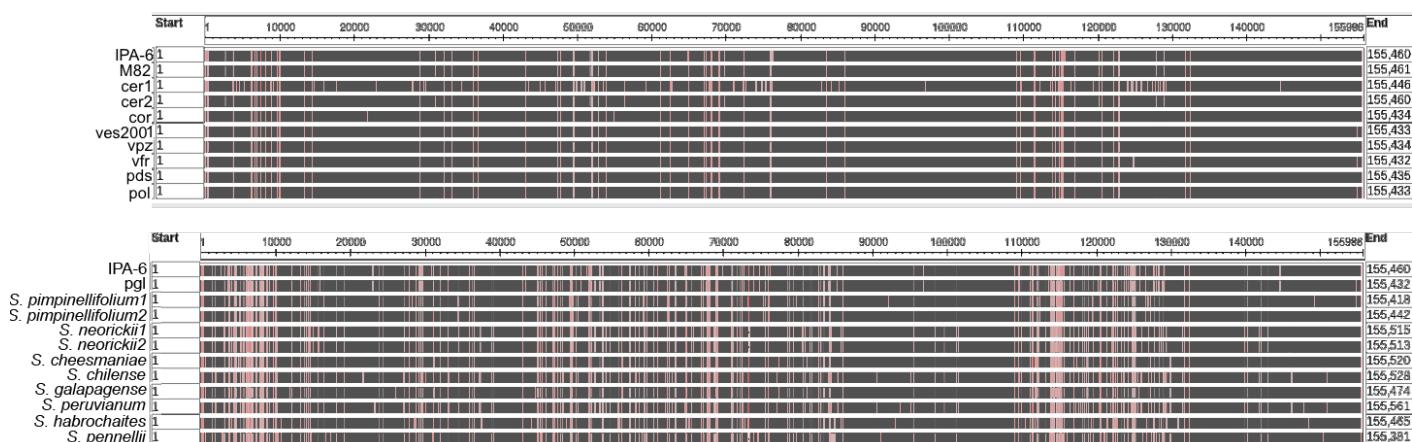


Figure S1. Overview of the nucleotide variability in nine plastomes sequenced in this study and in eleven species available in GenBank. The accession number AM087200 (cv. IPA-6) was used as reference. Red lines represent variable regions.

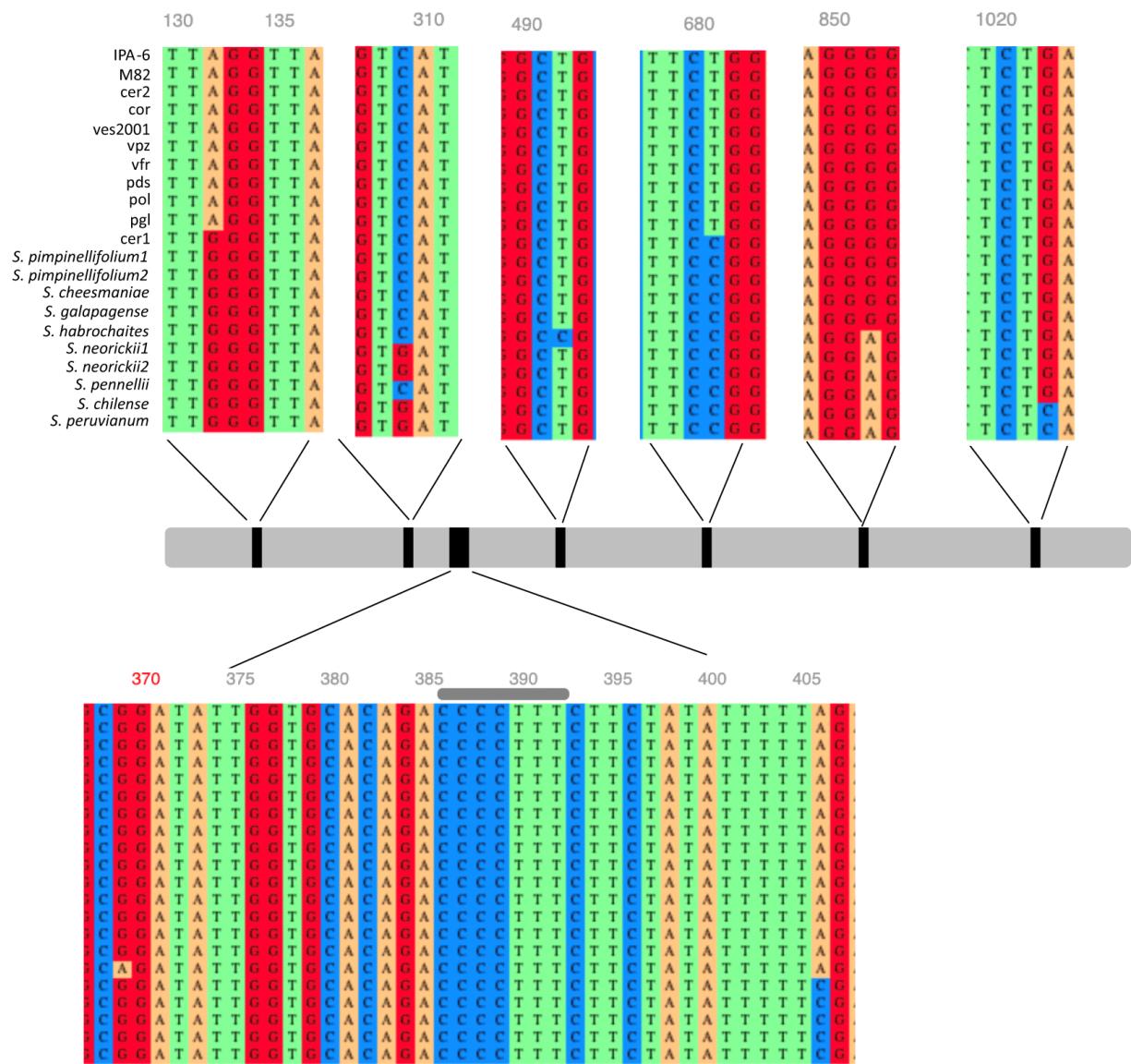


Figure S2. Schematic representation of the nucleotide variability observed in the *ndhH* gene for the plastomes under investigation. Grey bar represents the nucleotide multiple-sequence alignment (MSA) and it is scaled according to the MSA length. Black boxes indicate variable regions in the MSA. Above and below each box, a snapshot of the MSA along with alignment positions is reported.

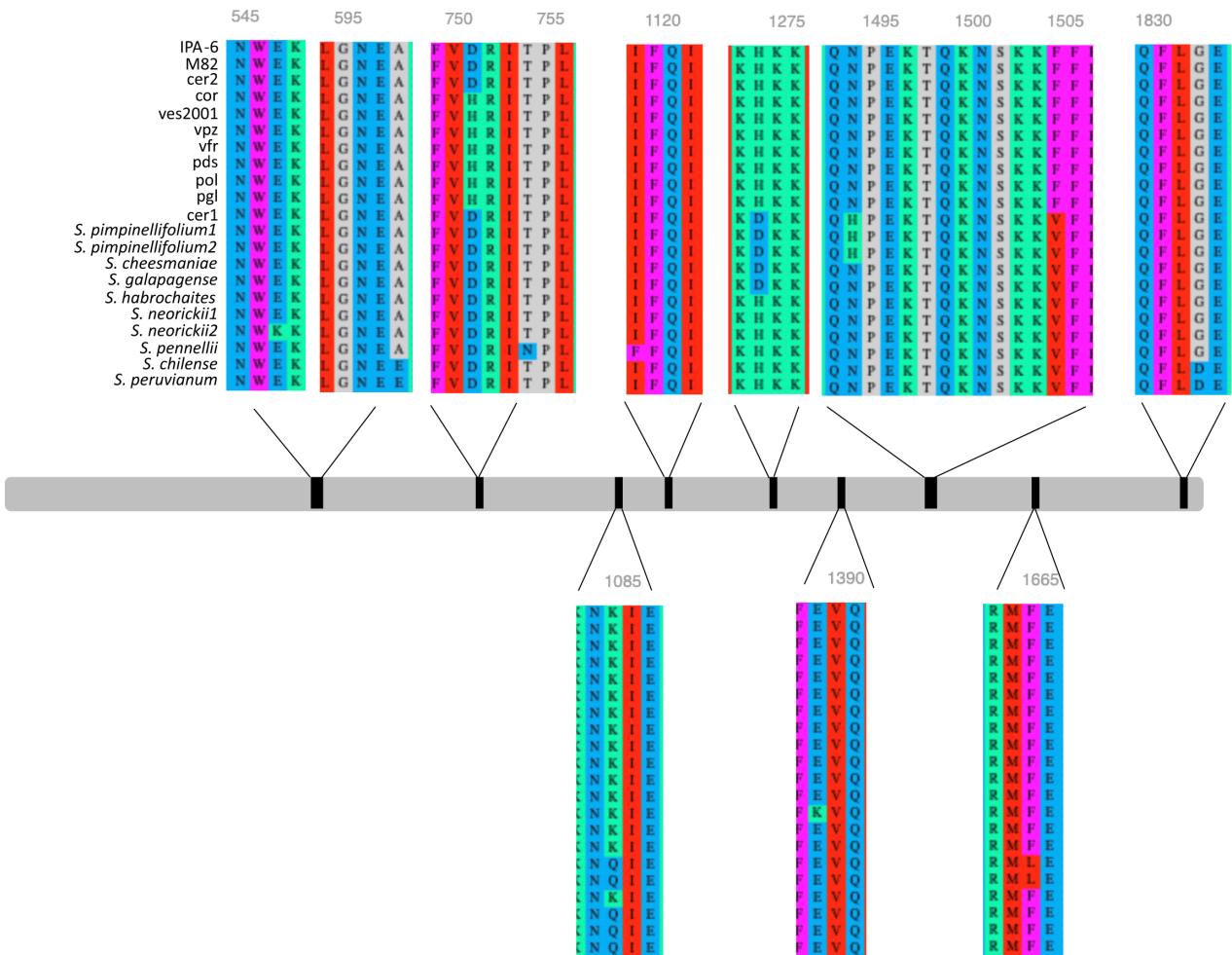
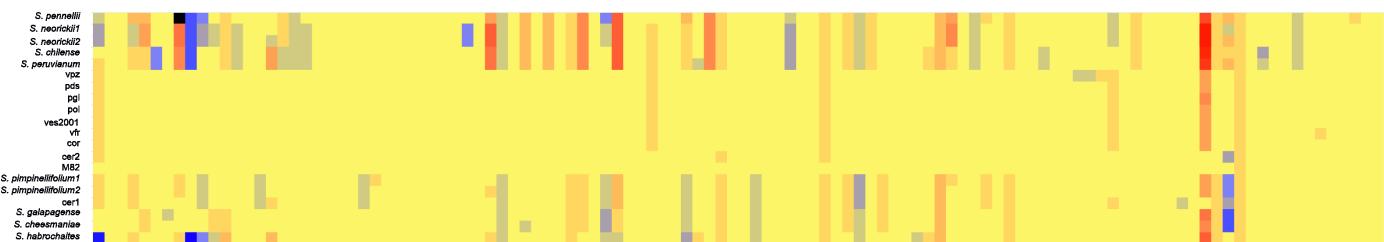


Figure S3. Schematic representation of the amino acid variability observed in the Ycf1 protein for the plastomes under investigation. Grey bar represents the amino acid multiple-sequence alignment (MSA) and it is scaled according to the MSA length. Black boxes indicate variable regions in the MSA. Above and below each box, a snapshot of the MSA along with alignment positions is reported.

a



b

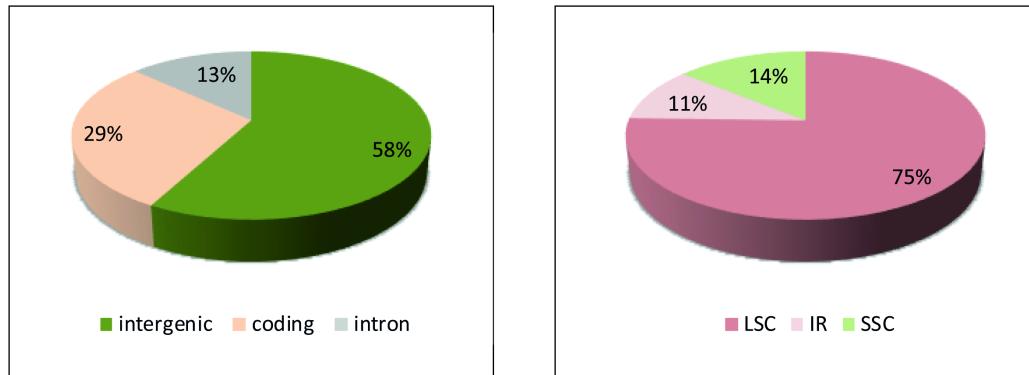
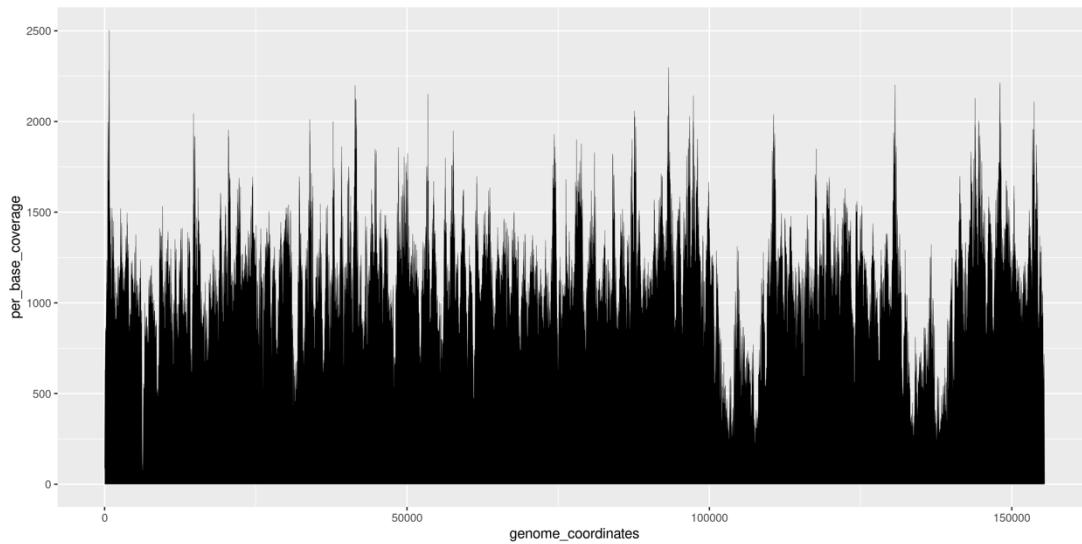


Figure S4. Simple sequence repeats (SSRs) in nine plastomes sequenced in this study and in eleven species available in GenBank. The plastome of IPA-6 (AM087200) was used as reference a) Heatmap representing differences in SSR size; colours range from red (SSR size larger than the reference) through yellow to blue (SSR size smaller than the reference). Black is for missing SSRs. b) Pie chart describing the percentage of SSRs located in coding sequences of genes, introns and intergenic regions as well as in the large single copy (LSC), small single copy (SSC) and inverted repeat b (IR) regions.

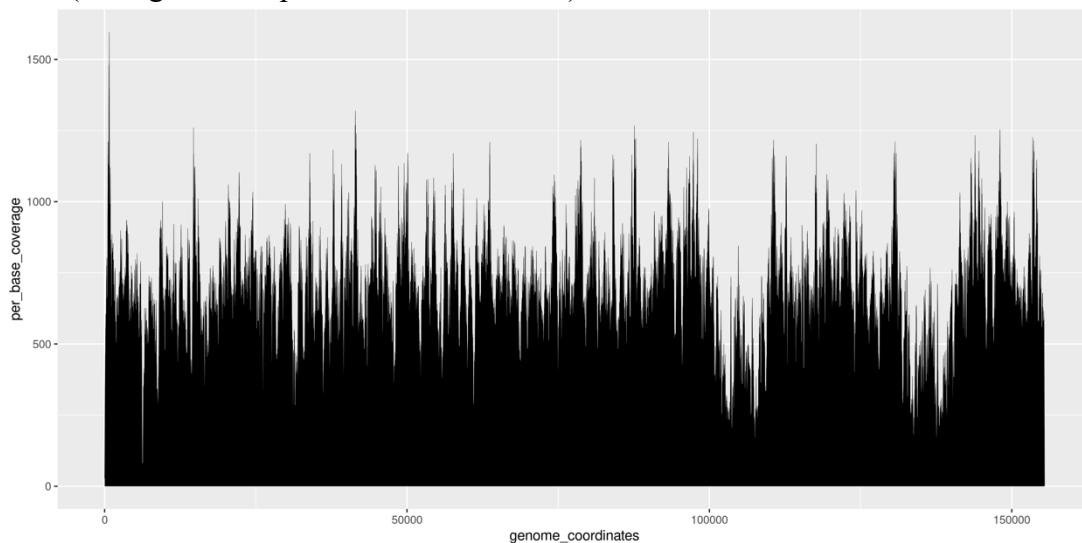
	114220	114230	114240
IPA-6	attggtcaaatgat	----	ataaaaaggatt
M82	attggtcaaatgat	----	ataaaaaggatt
cer1	attggtcaaatgat	----	ataaaaaggatt
cer2	attggtcaaatgat	----	ataaaaaggatt
cor	attggtcaaatgat	ataaaataaaaggatt	ataaaaaggatt
ves2001	attggtcaaatgat	ataaaataaaaggatt	ataaaaaggatt
vpz	attggtcaaatgat	ataaaataaaaggatt	ataaaaaggatt
vfr	attggtcaaatgat	ataaaataaaaggatt	ataaaaaggatt
pds	attggtcaaatgat	ataaaataaaaggatt	ataaaaaggatt
pol	attggtcaaatgat	ataaaataaaaggatt	ataaaaaggatt
pgl	attggtcaaatgat	ataaaataaaaggatt	ataaaaaggatt
<i>S. pimpinellifolium1</i>	attggtcaaatgat	----	ataaaaaggatt
<i>S. pimpinellifolium2</i>	attggtcaaatgat	----	ataaaaaggatt
<i>S. neorickii1</i>	attggtcaaatgat	----	ataaaaaggatt
<i>S. neorickii2</i>	attggtcaaatgat	----	ataaaaaggatt
<i>S. cheesmaniae</i>	attggtcaaatgat	----	ataaaaaggatt
<i>S. chilense</i>	attggtcaaatgat	----	ataaaaaggatt
<i>S. galapagense</i>	attggtcaaatgat	----	ataaaaaggatt
<i>S. peruvianum</i>	attggtcaaatgat	----	ataaaaaggatt
<i>S. habrochaites</i>	attggtcaaatgat	----	ataaaaaggatt
<i>S. pennellii</i>	attggtcaaatgat	----	ataaaaaggatt

Figure S5. Multiple-sequence alignment (MSA) of the region harbouring the duplicated sequence (ATAA)₂ scored only in local landraces.

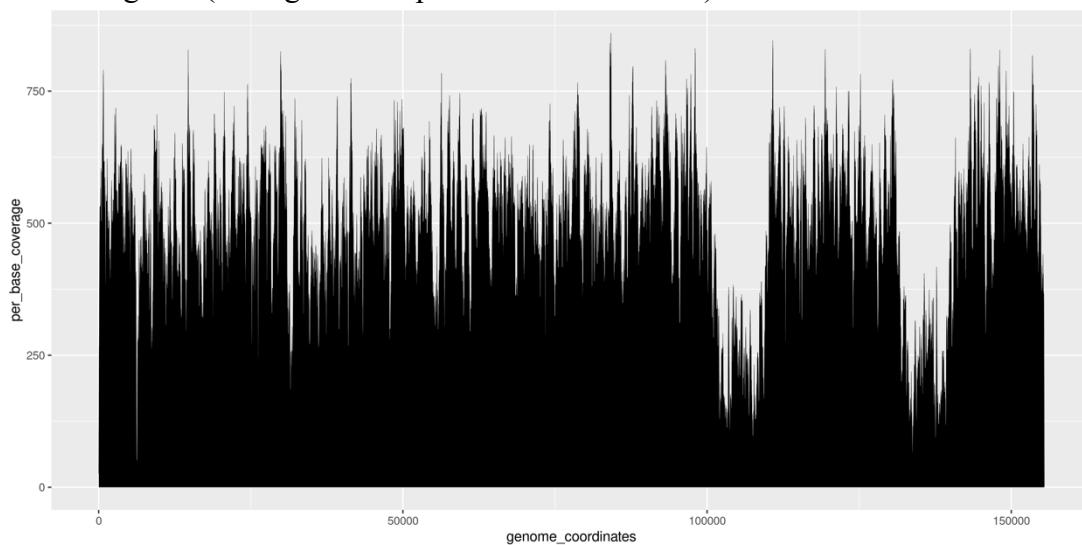
Corbarino (average read depth/nucleotide: 1072.84)



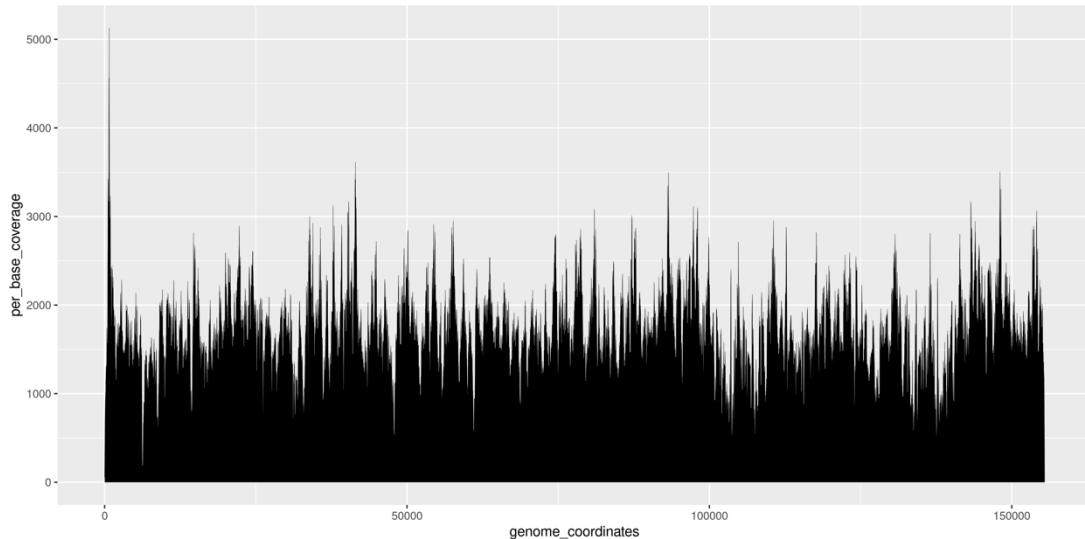
PDS (average read depth/nucleotide: 668.65)



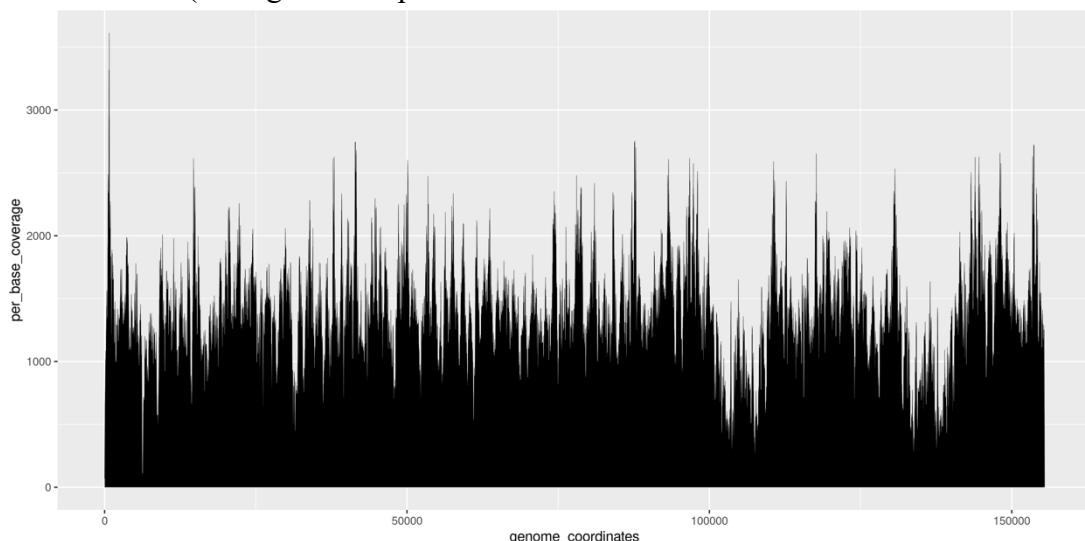
Piennolo giallo (average read depth/nucleotide: 476.94)



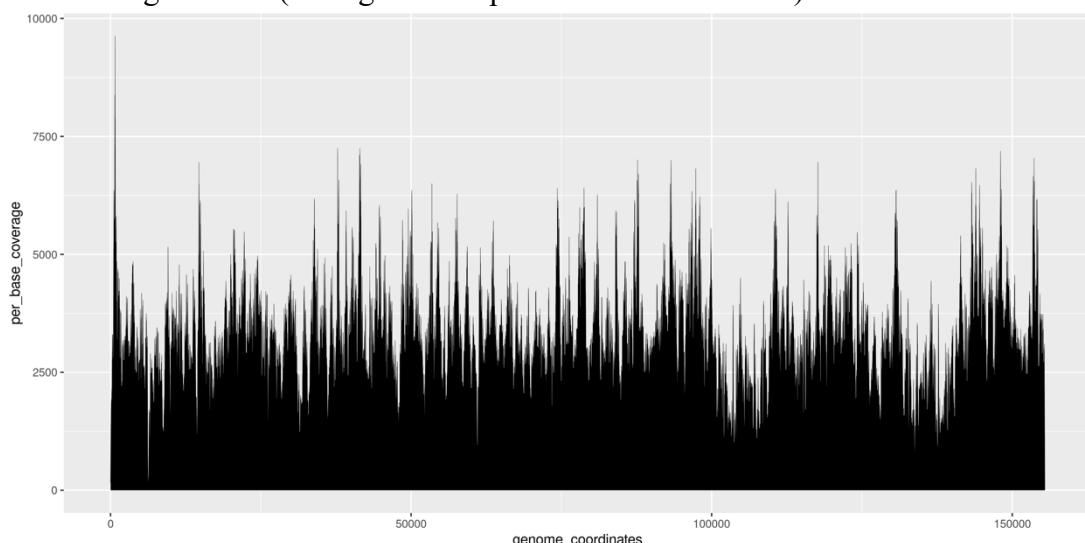
Pollena (average read depth/nucleotide: 1649.77)



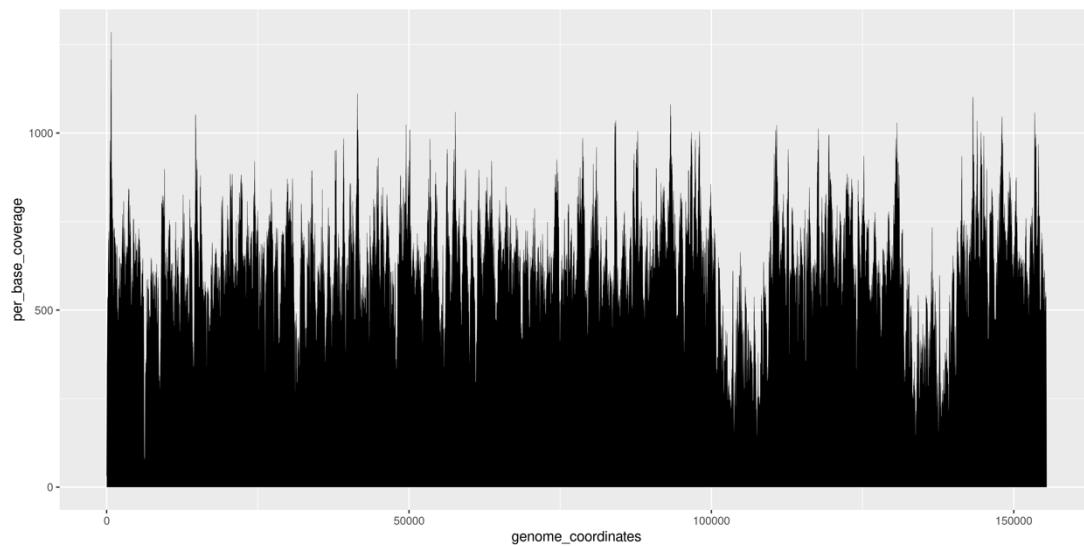
Vesuvio 2001 (average read depth/nucleotide: 1314.46)



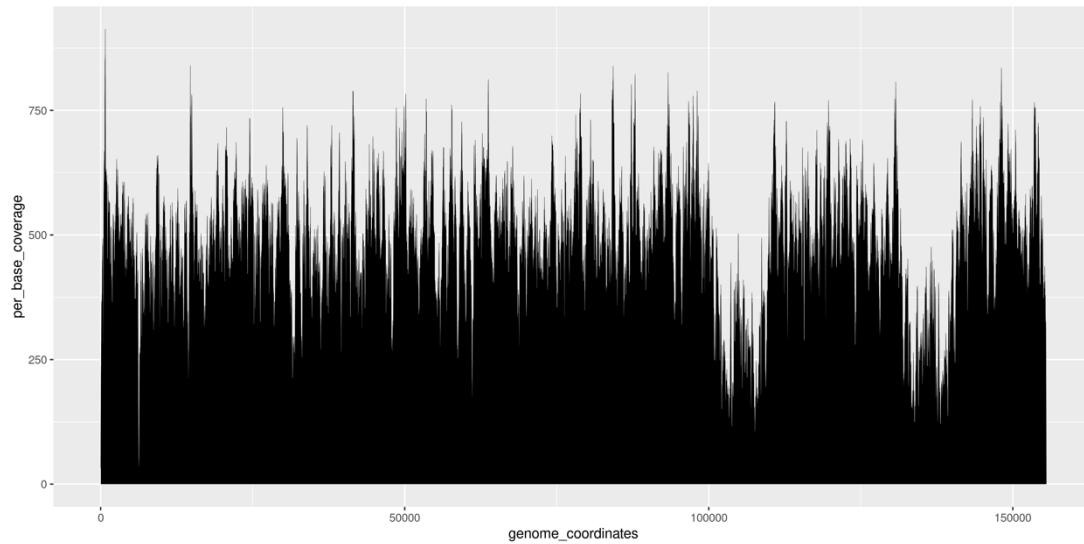
Vesuvio foglia riccia (average read depth/nucleotide: 3147.07)



Vesuviano pizzo (average read depth/nucleotide: 600.02)



S. neorickii 1 (average read depth/nucleotide: 456.53)



S. pimpinellifolium 1 (average read depth/nucleotide: 975.50)

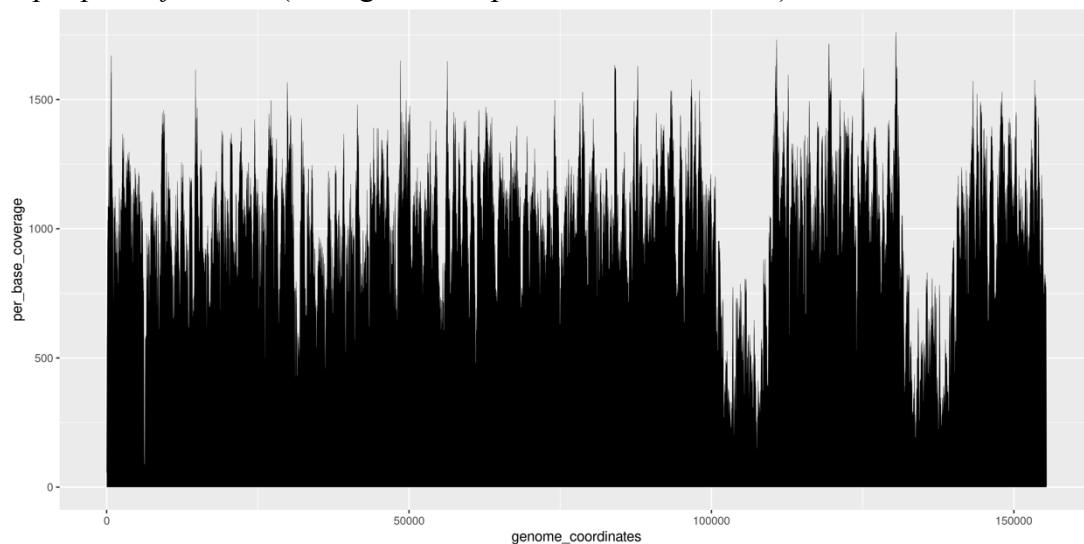


Figure S6. Distribution of per-base sequencing depth for each chloroplast genome sequenced in this work. The average coverage per-base is also reported.

Table S1. Tomato cpSSR primers developed in this study.

		LOCUS	Repeat	Sequence (5'-3')	Size (bp)	Fluorescent dye	Tm (°C)
LSC	intron	<i>tRNA-Lys (UUU)</i>	(T)12	GGTTATCAAATGATACTAGTGC GGAGATGTGTAGAAGAACAG	222	HEX	55
LSC	intron	<i>tRNA-Gly (UCC)</i>	(T)13	CTAGAAGGATCATCTACAAAG CGTTAGCTTGGAAAGGCTAGG	239	FAM	51
LSC	intergenic	<i>psbM - tRNA-Asp (GUC)</i>	(T)16	CGCCTTCTAAGAGGAGTAGGA CCGATGCATGTAATGGAATCG	314	ATTO550	59
LSC	intergenic	<i>atpB - rbcL</i>	(A)13	CATAGATTCTAGAGGAAATTCCA GCTTAGTCTCTGTTGTGG	389	FAM	55
LSC	intron	<i>clpP</i>	(T)17	CCAAACCGCGATGGTATTTCTT CAATTGGATTGGTAAGATATCTATG	288	ATTO565	58
LSC	intergenic	<i>rpl14 – rpl16</i>	(A)10	TGAGCATATCGACGATTGCT CCAAAATGCCTATACGAACTC	262	HEX	55
SSC	intergenic	<i>ndhF – rpl32</i>	(A)16-(A)19	TCATTCTACAGAACAGCCGAT TTCGAGCTAATAGTGTCTCC	258-261	ATTO550	59
SSC	intergenic	<i>rpl32 - tRNA-Leu (UAG)</i>	(A)15	AGACCCGCCATGTATATCTTGC CTGAGTCTTCCTATATTTCTG	292	ATTO565	58

Table S2. Simple Sequence Repeats (SSRs) in the twenty-one tomato chloroplast genomes using IPA-6 (AM087200) as reference genome. SSRs size, location and distribution among different regions, namely coding, intron, intergenic are reported. The *unit_size/min_repeats* parameters were as follows: 1/8, 2/6, 3/5, 4/5, 5/5, 6/5. SSRs located in IRa were not counted. SSRs were identified using MISA-MicroSatellite identification tool (<http://pgrc.ipk-gatersleben.de/misa/>).

	location	microsatellite	IPA-6	M82	cor	pds	pgl	pol	vpz	ves2001	vfr	cer1	cer2	S. pimpinellifolium 1	S. pimpinellifolium 2	S. neorickei 1	S. neorickei 2	S. habrocaltes	S. peruvianum	S. cheesmaniae	S. galapagense	S. pennelli	S. chilense
LSC intergenic	tRNA-His(GUG)-psBA	(A)n	13	13	12	12	12	12	12	12	12	12	12	12	15	15	15	18	12	13	13	14	13
LSC coding	matK	(T)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC coding	matK	(T)n	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	
LSC intron	tRNA-Lys(UUU)	(T)n	12	12	12	12	12	12	12	12	12	11	12	11	11	13	11	11	12	12	10	11	
LSC intergenic	tRNA-Lys(UUU)-rps16	(A)n	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	10	10	10	9	10	
LSC intergenic	rps16 - tRNA-Gln(UUG)	(AT)n	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	7	4	4	7	
LSC intergenic	rps16 - tRNA-Gln(UUG)	(T)n	7	7	7	7	7	7	7	7	7	7	7	7	7	7	7	7	7	7	7	7	
LSC intergenic	rps16 - tRNA-Gln(UUG)	(T)n	16	16	16	16	16	16	16	16	16	16	16	15	15	11	11	15	12	16	-	12	
LSC intergenic	rps16 - tRNA-Gln(UUG)	(T)n	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	13	13	
LSC intergenic	psbK - psbA	(A)n	11	11	11	11	11	11	11	11	11	12	11	12	12	13	13	14	11	11	11	14	11
LSC intergenic	psbK - psbA	(T)n	11	11	11	11	11	11	11	11	11	11	11	11	11	11	12	12	11	10	10	11	11
LSC intergenic	psbI - tRNA-Ser(GCU)	(A)n	11	11	11	11	11	11	11	11	11	11	11	11	11	11	10	10	9	10	10	10	10
LSC intergenic	tRNA-Ser(GCU) - tRNA-Gly(UCC)	(T)n	7	7	7	7	7	7	7	7	7	7	7	7	7	8	8	7	8	7	7	8	
LSC intergenic	tRNA-Ser(GCU) - tRNA-Gly(UCC)	(A)n	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	
LSC intergenic	tRNA-Ser(GCU) - tRNA-Gly(UCC)	(A)n	8	8	8	8	8	8	8	8	8	8	9	8	9	8	8	8	8	8	8	8	
LSC intron	tRNA-Gly(UCC)	(T)n	13	13	13	13	13	13	13	13	13	13	12	13	13	13	14	11	10	13	13	13	10
LSC intergenic	tRNA-Gly(UCC) - tRNA-Arg(UCU)	(T)n	12	12	12	12	12	12	12	12	12	12	12	12	12	11	11	12	13	12	12	11	13
LSC intergenic	tRNA-Gly(UCC) - tRNA-Arg(UCU)	(A)n	7	7	7	7	7	7	7	7	7	7	7	7	7	8	7	8	7	7	8	8	
LSC intergenic	tRNA-Arg(UCU) - atpA	(T)n	9	9	9	9	9	9	9	9	9	9	9	9	9	10	10	9	10	9	9	9	10
LSC intergenic	atpA - atpF	(A)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
LSC intron	atpF	(T)n	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9
LSC coding	atpF	(A)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
LSC intergenic	atpH - atpI	(A)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
LSC intergenic	atpH - atpI	(T)n	8	8	8	8	8	8	8	8	8	8	9	8	9	8	8	8	8	8	8	8	8
LSC coding	rps2 - rpoC2	(A)n	11	11	11	11	11	11	11	11	11	11	11	11	10	11	11	11	11	11	11	11	11
LSC coding	rpoC2	(T)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
LSC coding	rpoC2	(C)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
LSC coding	rpoC2	(T)n	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
LSC coding	rpoC1	(A)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
LSC coding	rpoC1	(T)n	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11
LSC coding	rpoB	(T)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8
LSC intergenic	tRNA-Cys(GCA) - petN	(TA)n	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
LSC intergenic	petN - psbM	(A)n	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11
LSC intergenic	psbM - tRNA-Asp(GUC)	(T)n	16	16	16	16	16	16	16	16	16	16	16	16	16	15	10	15	11	16	13	11	
LSC intergenic	tRNA-Glu(UUC) - tRNA-Thr(GGU)	(T)n	8	8	8	8	8	8	8	8	8	8	9	8	9	9	9	9	9	9	9	9	
LSC intergenic	tRNA-Thr(GGU) - psbD	(T)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC intergenic	psbC - tRNA-Ser(UGA)	(T)n	12	12	12	12	12	12	12	12	12	12	12	12	12	10	10	12	10	13	12	10	
LSC intron	ycf3	(T)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC intron	ycf3	(A)n	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11	11
LSC intergenic	ycf3 - tRNA-Ser(GGA)	(T)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC intergenic	ycf3 - tRNA-Ser(GGA)	(A)n	8	8	8	8	8	8	8	8	8	8	7	8	7	7	7	7	7	7	7	7	
LSC intergenic	rps4 - tRNA-Thr(UGU)	(T)n	10	10	10	10	10	10	10	10	10	10	9	10	9	9	6	10	6	9	6	6	
LSC intergenic	rps4 - tRNA-Thr(UGU)	(A)n	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	
LSC intergenic	rps4 - tRNA-Thr(UGU)	(A)n	10	10	10	10	10	10	10	10	10	10	11	10	11	11	11	11	11	12	12	13	
LSC intergenic	tRNA-Thr(UGU) - tRNA-Leu(UAA)	(T)n	11	11	11	11	11	11	11	11	11	9	11	9	9	5	5	12	5	10	10	5	
LSC intergenic	tRNA-Leu(UAA) - tRNA-Phe(GAA)	(T)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC coding	ndhC	(A)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC intergenic	ndhC - tRNA-Val(UAC)	(T)n	10	10	9	9	9	9	9	9	9	9	10	10	10	10	9	9	10	9	10	9	
LSC intron	tRNA-Val(UAC)	(T)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC intron	tRNA-Val(UAC)	(A)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC intergenic	tRNA-Met(CAU) - atpE	(T)n	12	12	12	12	12	12	12	12	12	13	12	13	13	11	11	14	11	12	12	13	
LSC coding	atpB	(T)n	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
LSC intergenic	atpB - rbcL	(A)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC intergenic	rbcL - accD	(T)n	13	13	13	13	13	13	13	13	13	13	12	12	12	12	12	12	12	12	12	12	12
LSC intergenic	accD - psdA	(T)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC intergenic	psdA - ycf4	(T)n	10	10	10	10	10	10	10	10	10	10	11	10	11	11	10	11	10	11	11	10	10
LSC coding	ycf4	(T)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC intergenic	ycf4 - ycf10	(T)n	10	10	10	10	10	10	10	10	10	10	10	10	10	10	12	10	10	10	11	12	
LSC intergenic	ycf4 - ycf10	(A)n	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	9	
LSC coding	psbF	(A)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC intergenic	psbE - petL	(T)n	9	9	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC intergenic	petL - petG	(T)n	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	8	
LSC intergenic	petL - petG	(A)n	8	8	8	8	8	8	8	8	8	8	8	8	8	10	9	9	9	9	9	9	
LSC intergenic	tRNA-Pro(UGG) - psaJ	(T)n	10	10	10	10	10	10	10	10	10	10	10	10	10	9	9	9	10	9	9	9	
LSC intergenic																							

Table S3. Tandem Repeats (TRs) in the twenty-one tomato chloroplast genomes using IPA-6 (AM087200) as reference genome. TRs copy number and location are reported. TRs were identified using the tool available at <https://tandem.bu.edu/trf/trf.basic.submit.html>.

Sequence	Period	Copy Number																			location			
		Size	IPA-6	M82	cor	pds	pgl	pol	vpz	ves2001	vfr	cer1	cer2	<i>S. pimpinellifolium</i> 1	<i>S. pimpinellifolium</i> 2	<i>S. neorickii</i> 1	<i>S. neorickii</i> 2	<i>S. habrocaites</i>	<i>S. peruvianum</i>	<i>S. cheesmaniae</i>	<i>S. galapagense</i>	<i>S. pennelli</i>	<i>S. chilense</i>	
GTTCAATGATAATTATTATT	19	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2.0	LSC <i>tRNA-His (GUG)-psbA</i>	intergenic	
ATCATGAATAAT	13	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	-	-	2.0	-	2.0	2.0	-	-	LSC <i>tRNA-His (GUG)-psbA</i>	intergenic
TATTTCCTTAATT	13	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	LSC <i>rps16</i>	genic	
TAATACATAGAT	13	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	LSC <i>rps16-tRNA-Gln (UUG)</i>	intergenic	
ACTTACITTCCTTCCCTTATA	22	2.0	2.0	-	-	-	-	-	-	-	-	-	-	-	2.0	3.0	3.0	2.0	2.0	2.0	2.0	LSC <i>tRNA-Gln (UUG)-psbK</i>	intergenic	
GGTTAAAGCTGA	13	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	LSC <i>rps4</i>	genic	
ACGTAATACATATTAGAAAG	20	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2.0	2.0	-	-	2.0	2.0	LSC <i>tRNA-Phe (GAA)-ndhJ</i>	intergenic	
AGTTAAATAGTTTACCC	17	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	LSC <i>PsbE-petL</i>	intergenic	
TTTTTCACTCTAGITTA	17	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2.1	2.1	-	-	-	-	LSC <i>psaI-rpl33</i>	intergenic	
GAATCTAGATTTGAGT	16	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	LSC <i>rps18-rpl20</i>	intergenic	
CATAGATATCTTA	13	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	LSC <i>clpP</i>	intronic	
AGAAATTAAGAAATTAGAA	17	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	2.1	-	-	2.1	2.1	-	-	LSC <i>rpl16-rps3</i>	intergenic	
AATCAATGCAATTAGAGG	20	3.4	3.4	3.4	3.4	3.4	3.4	3.4	3.4	3.4	3.4	3.4	3.4	3.4	3.4	3.4	3.4	3.4	3.4	3.4	3.4	Irb <i>tRNA-Ile -ycf2</i>	intergenic	
ACTTAGTGACTCTTCTTC	20	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	Irb <i>rps12-tRNA-Val (GAC)</i>	intergenic	
GTCATTAATATCCTTG	16	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2.0	Irb <i>tRNA-Arg (ACG)-tRNA-Asn (GUU)</i>	intergenic	
AGAAGAACGTATAAAA	18	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2.1	2.1	-	-	-	-	SSC <i>rpl32-tRNA-Leu (UAG)</i>	intergenic	
TGTTTTAGATCCGAACTAAGCCITC	26	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2.0	2.0	SSC <i>ndhG-ndhI</i>	intergenic