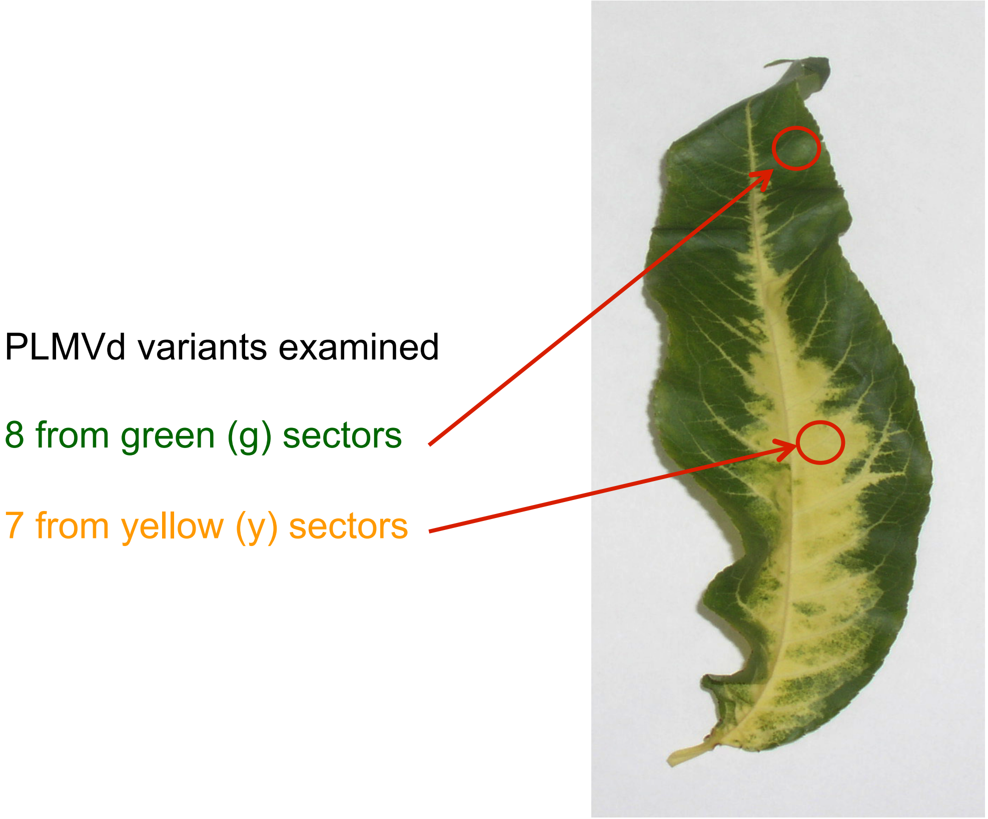
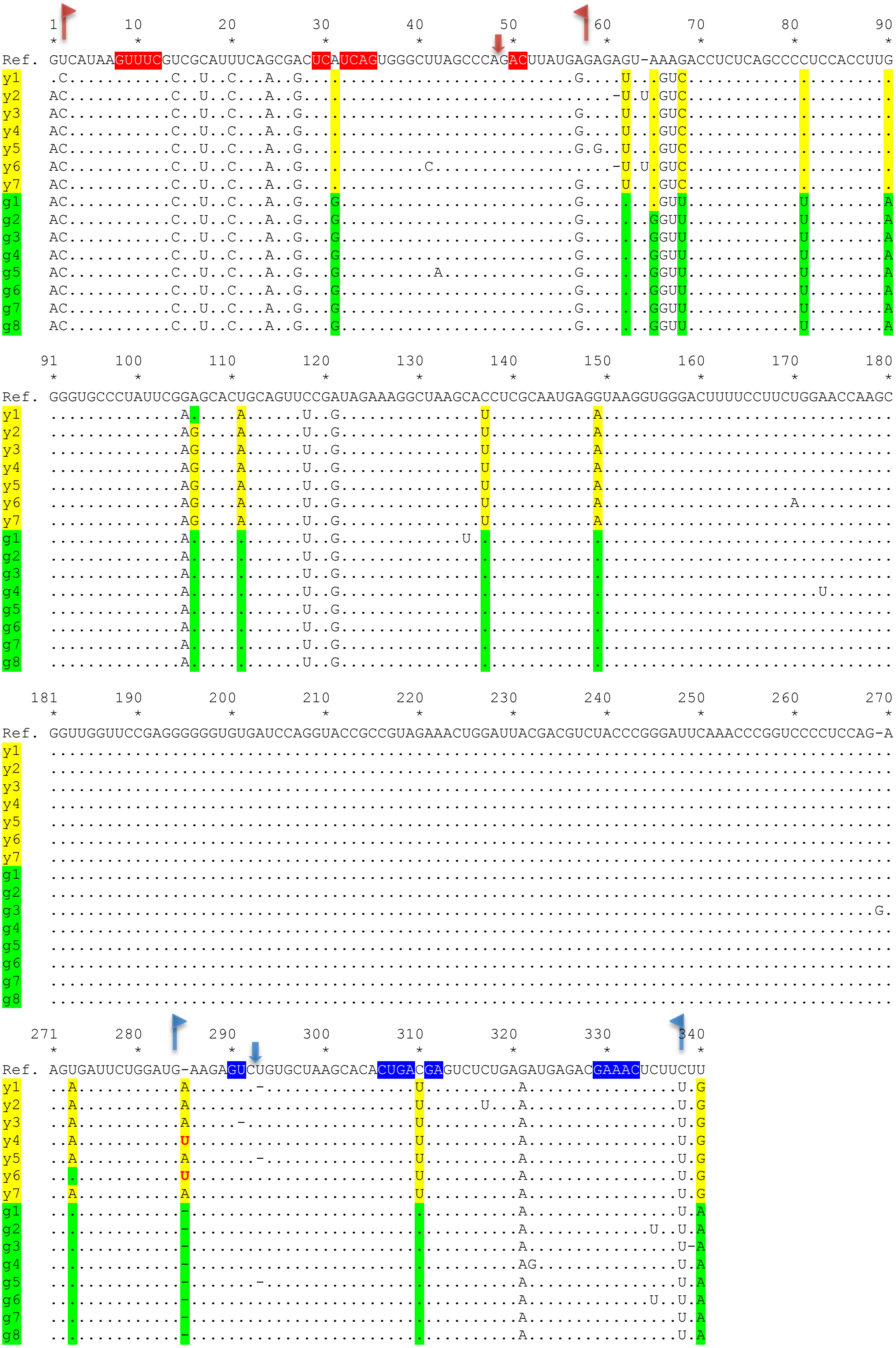


Figure S1. Scheme of the approach used to dissect PLMVd populations from a PYM-expressing leaf of peach cv. "O'Henry". Nucleic acid preparations from green (g) and yellow (y) sectors were subjected to RT-PCR using the PLMVd-specific pair of primers RF43 and RF44, with the resulting full-length products being cloned and sequenced.



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     1        10        20        30        40        50        60        70        80        90
     *        *         *         *         *         *         *         *         *         *
Ref. GUCAUAAGUUUCGUCGCAUUUCAGCGACUCAUCAGUGGGCUUAGCCCAGACUUAUGAGAGAGU-AAAGACCUCUCAGCCCCUCCACCUUG
y1   .C...........C..U..C...A..G.............................G....U...GUC......................
y2   AC...........C..U..C...A..G.................................-U.U.GUC......................
y3   AC...........C..U..C...A..G.............................G....U...GUC......................
y4   AC...........C..U..C...A..G.............................G....U...GUC......................
y5   AC...........C..U..C...A..G.............................G.G..U...GUC......................
y6   AC...........C..U..C...A..G.............C...................-U.U.GUC......................
y7   AC...........C..U..C...A..G.............................G....U...GUC......................
g1   AC...........C..U..C...A..G...G.........................G........GUU............U........A
g2   AC...........C..U..C...A..G...G.........................G.......GGUU............U........A
g3   AC...........C..U..C...A..G...G.........................G.......GGUU............U........A
g4   AC...........C..U..C...A..G...G.........................G.......GGUU............U........A
g5   AC...........C..U..C...A..G...G..........A..............G.......GGUU............U........A
g6   AC...........C..U..C...A..G...G.........................G.......GGUU............U........A
g7   AC...........C..U..C...A..G...G.........................G.......GGUU............U........A
g8   AC...........C..U..C...A..G...G.........................G.......GGUU............U........A

    91      100       110       120       130       140       150       160       170       180
     *        *         *         *         *         *         *         *         *         *
Ref. GGGUGCCCUAUUCGGAGCACUGCAGUUCCGAUAGAAAGGCUAAGCACCUCGCAAUGAGGUAAGGUGGGACUUUUCCUUCUGGAACCAAGC
y1   ..............A.....A......U..G...............U...........A...............................
y2   ..............AG....A......U..G...............U...........A...............................
y3   ..............AG....A......U..G...............U...........A...............................
y4   ..............AG....A......U..G...............U...........A...............................
y5   ..............AG....A......U..G...............U...........A...............................
y6   ..............AG....A......U..G...............U...........A....................A..........
y7   ..............AG....A......U..G...............U...........A...............................
g1   ..............A............U..G.............U.............................................
g2   ..............A............U..G...........................................................
g3   ..............A............U..G...........................................................
g4   ..............A............U..G...................................................U.......
g5   ..............A............U..G...........................................................
g6   ..............A............U..G...........................................................
g7   ..............A............U..G...........................................................
g8   ..............A............U..G...........................................................

   181      190       200       210       220       230       240       250       260       270
     *        *         *         *         *         *         *         *         *         *
Ref. GGUUGGUUCCGAGGGGGGUGUGAUCCAGGUACCGCCGUAGAAACUGGAUUACGACGUCUACCCGGGAUUCAAACCCGGUCCCCUCCAG-A
y1   ..........................................................................................
y2   ..........................................................................................
y3   ..........................................................................................
y4   ..........................................................................................
y5   ..........................................................................................
y6   ..........................................................................................
y7   ..........................................................................................
g1   ..........................................................................................
g2   ..........................................................................................
g3   ........................................................................................G.
g4   ..........................................................................................
g5   ..........................................................................................
g6   ..........................................................................................
g7   ..........................................................................................
g8   ..........................................................................................

   271      280       290       300       310       320       330       340
     *        *         *         *         *         *         *         *
Ref. AGUGAUUCUGGAUG-AAGAGUCUGUGCUAAGCACACUGACGAGUCUCUGAGAUGAGACGAAACUCUUCUU
y1   ..A...........A.......-................U..........A................U.G
y2   ..A...........A........................U......U...A................U.G
y3   ..A...........A.....-..................U..........A................U.G
y4   ..A...........U........................U..........A................U.G
y5   ..A...........A.......-................U..........A................U.G
y6   ..............U........................U..........A................U.G
y7   ..A...........A........................U..........A................U.G
g1   ..............-...................................A................U.A
g2   ..............-...................................A.............U..U.A
g3   ..............-...................................A................U-A
g4   ..............-...................................AG...............U.A
g5   ..............-.......-...........................A................U.A
g6   ..............-...................................A.............U..U.A
g7   ..............-...................................A................U.A
g8   ..............-...................................A................U.A
```

Figure S2. Multiple alignment of PLMVd variants obtained from the original PYM source. Nucleic acid preparations from yellow and green leaf sectors were subjected to RT-PCR with the PLMVd-specific pair of primers RF43 and RF44 (Ambrós et al., 1998). For comparative purposes the reference variant of PLMVd —GenBank M83545.1 with two minor corrections (Ambrós et al., 1998)— appears on top. Dots indicate nucleotides identical to the reference sequence, and dashes gaps. Regions forming the plus and minus hammerhead structures are delimited by flags, the nucleotides conserved in most natural hammerhead structures are on colored backgrounds, and the self-cleavage sites are shown by arrows; blue and red symbols refer to plus and minus polarities, respectively. Nucleotides characteristic of PLMVd variants retrieved from yellow (y) and green (g) leaf sectors are indicated with the corresponding colors. Notice that the U283 of variant PYM-y4 corresponds to position 285 in the multiple alignment due to the existence of indels.

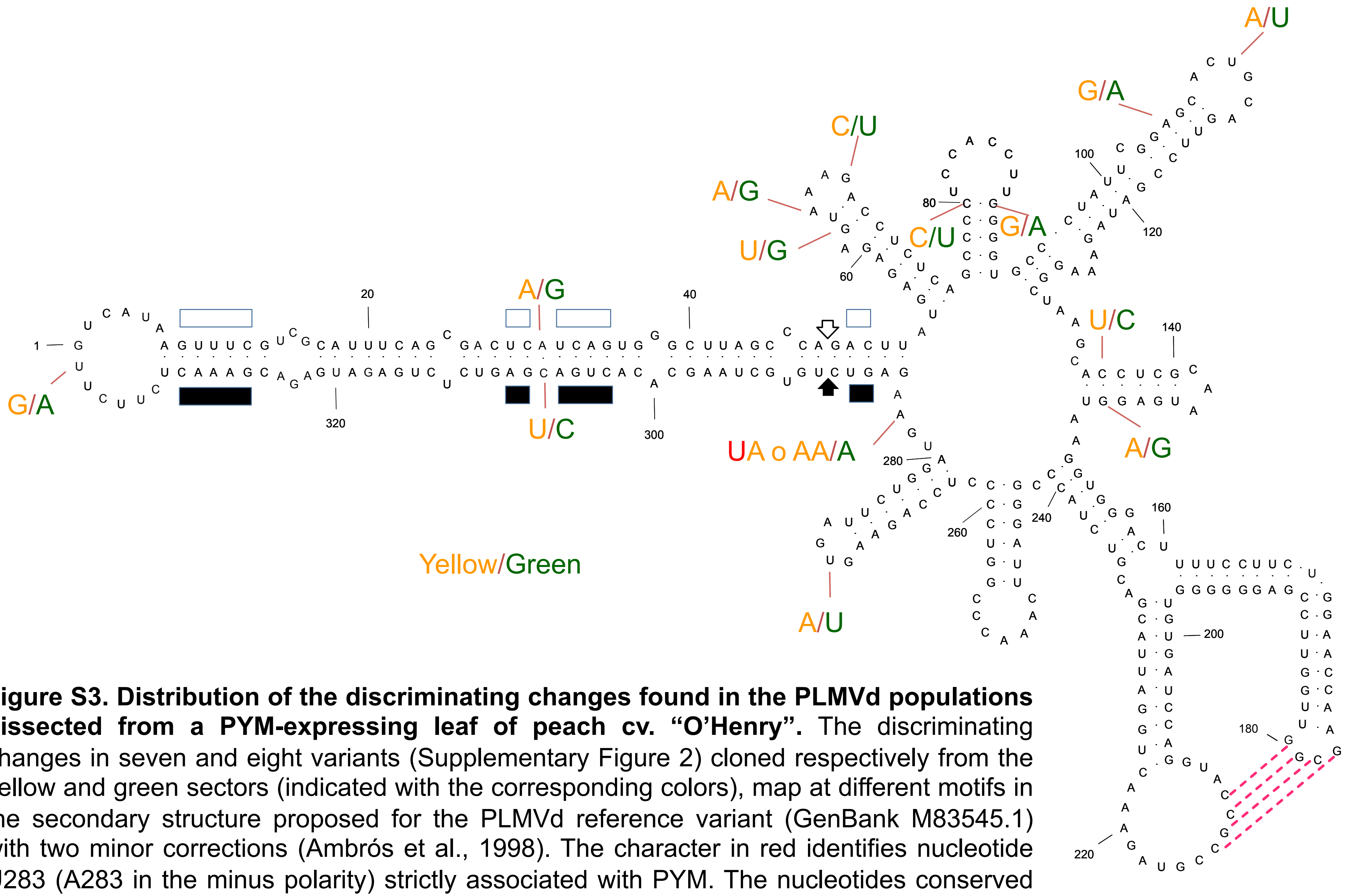

Figure S3. Distribution of the discriminating changes found in the PLMVd populations dissected from a PYM-expressing leaf of peach cv. "O'Henry". The discriminating changes in seven and eight variants (Supplementary Figure 2) cloned respectively from the yellow and green sectors (indicated with the corresponding colors), map at different motifs in the secondary structure proposed for the PLMVd reference variant (GenBank M83545.1) with two minor corrections (Ambrós et al., 1998). The character in red identifies nucleotide U283 (A283 in the minus polarity) strictly associated with PYM. The nucleotides conserved in most natural hammerhead structures are denoted with rectangles, and the self-cleavage sites by arrows; black and white symbols refer to (+) and (-) polarities, respectively. Residues involved in a kissing-loop interaction are indicated by broken red lines.

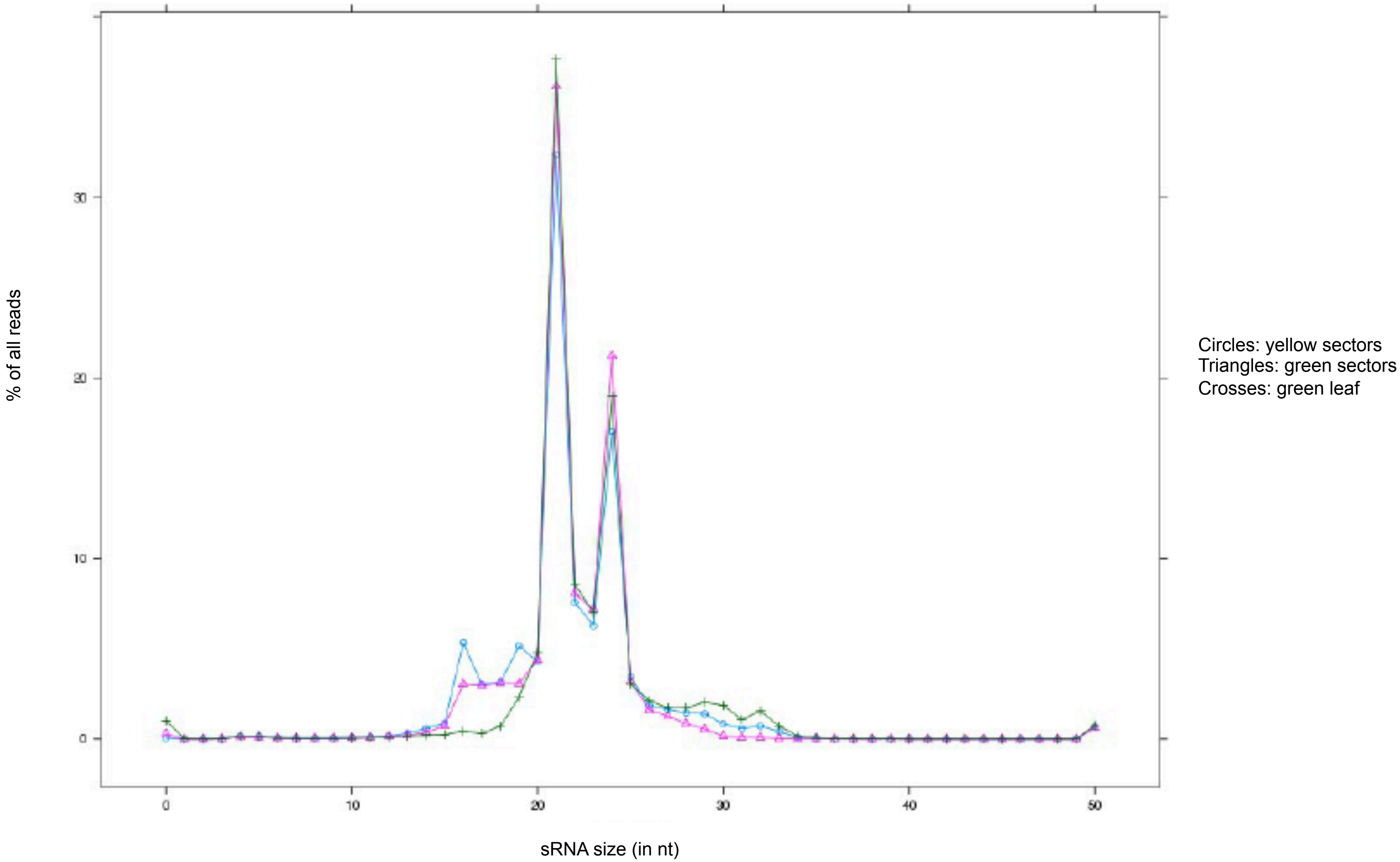

Figure S4. Relative distribution by size classes of total sRNA reads from a GF-305 peach seedling inoculated with PYM-variant y4. Nucleic acid preparations were obtained from yellow (circles) and green (triangles) sectors of a PYM-expressing leaf, and of a fully green leaf (crosses) of the same plant. Two main 21- and 24-nt peaks are observed in the three samples.

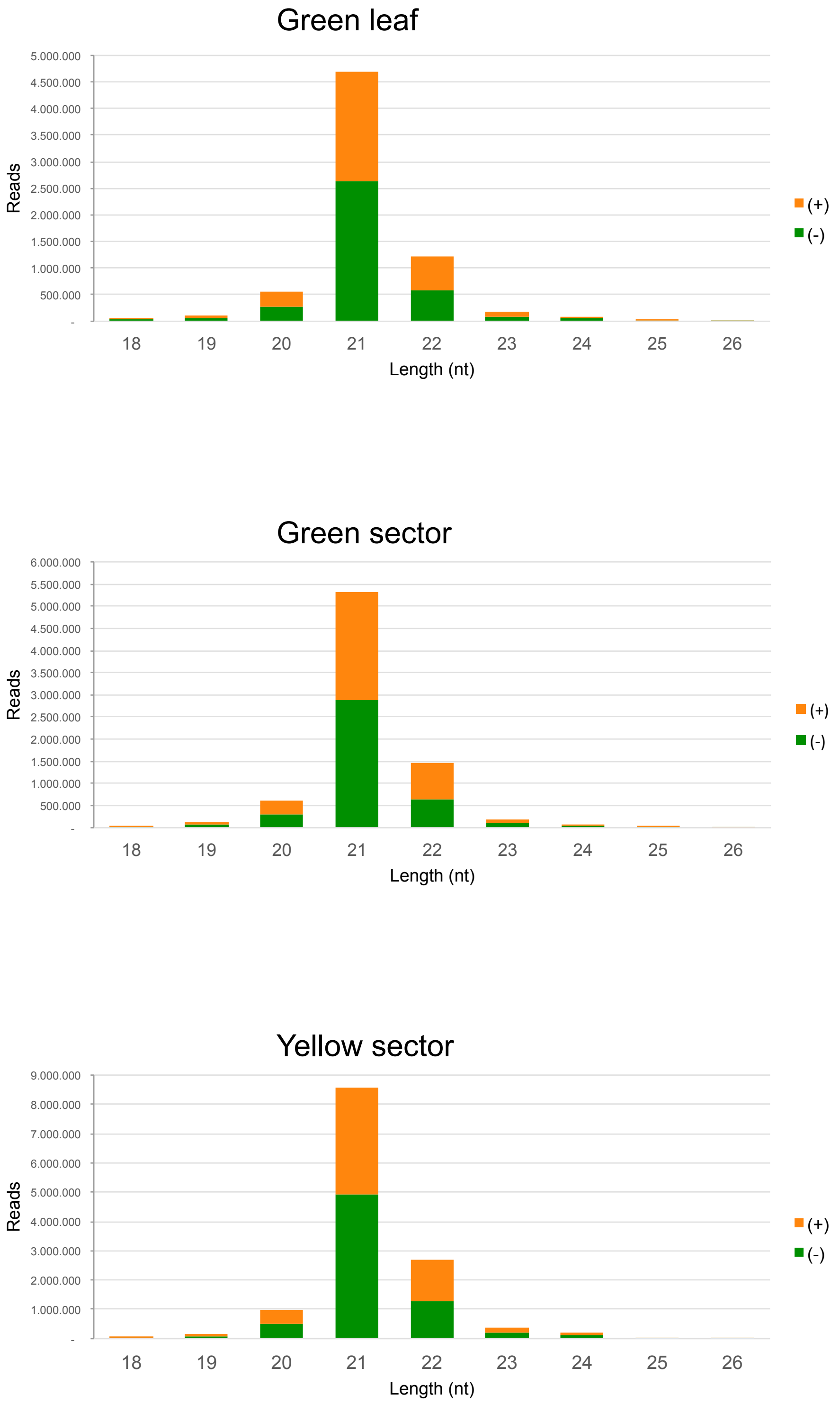

Figure S5. Size distribution of PLMVd sRNA reads from a GF-305 peach seedling inoculated with PYM-variant y4. The histograms compare (+) and (-) reads of 18 to 26 nt in nucleic acid preparations from yellow and green sectors of a PYM-expressing leaf, and from a fully green leaf of the same plant.

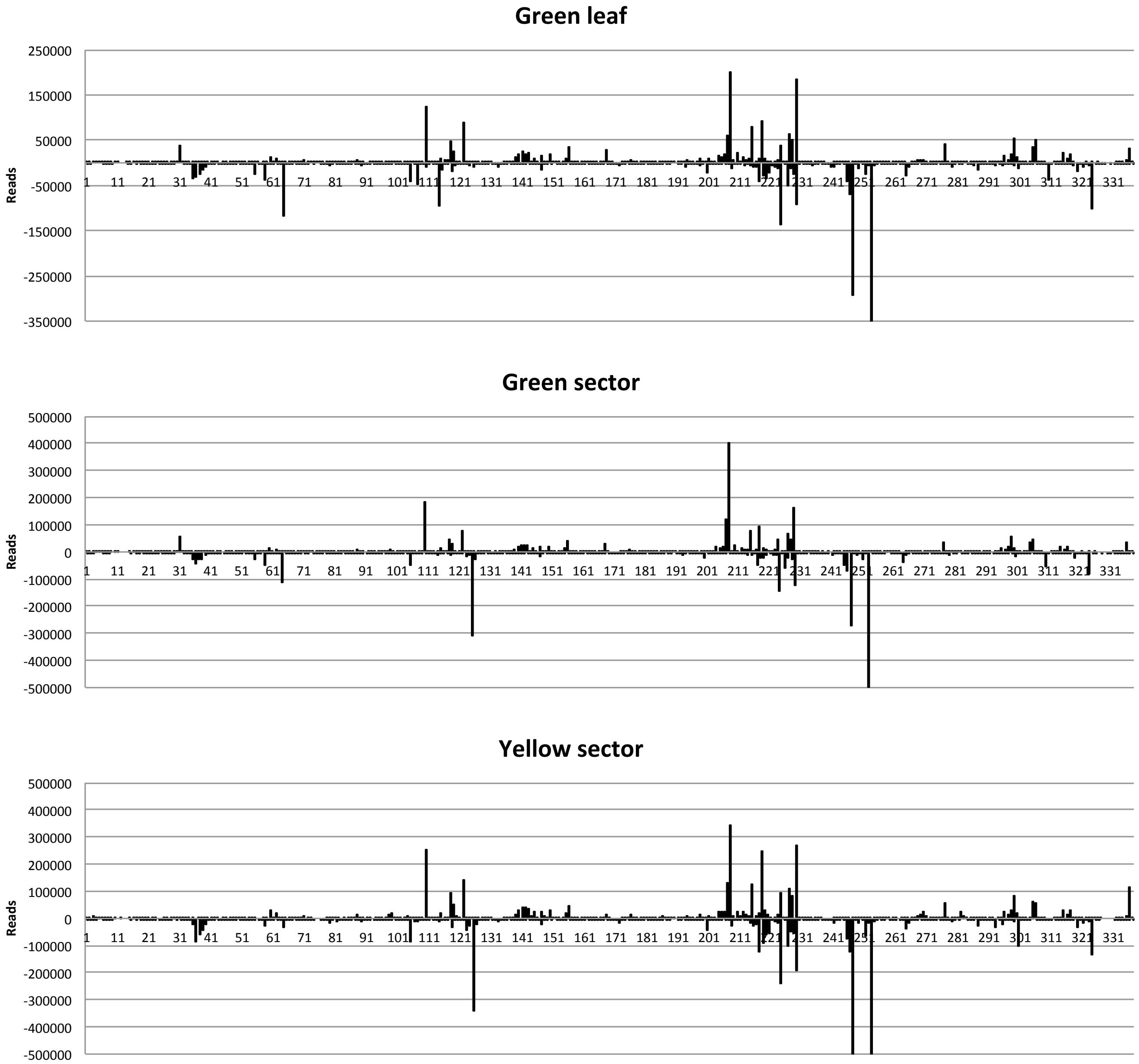

Figure S6. Hot spot profiles of PLMVd sRNA reads from a GF-305 peach seedling inoculated with PYM-variant y4. Location and frequency in the genomic PLMVd RNA of the 5' termini of the (+) (upper panels) and (-) (lower panels) PLMVd sRNA reads in nucleic acid preparations from yellow and green sectors of a PYM-expressing leaf, and from a fully green leaf of the same plant. Note that the same numbers are used in the (+) strand (5'→3' orientation is from left to right) and in the (-) strand (5'→3' orientation is from right to left).

Table S1. RNA duplexes formed between the cpSecA mRNA and PLMVd-sRNAs derived from the same region as PYM-sRNA40 in different PLMVd variants

PLMVd variant	cpSecA mRNA:PLMVd-sRNA duplex[a]	Symptoms

y4 (MK212036)	`5'- CUGAAGAGAUUCUGGGUGUAA-3'`
	`    |-|||||||||||||o|||||`
	`3'- GUCUUCUCUAAGACCUACAUU-5'`
		peach yellow mosaic

y7	`5'- CUGAAGAGAUUCUGGGUGUAA-3'`
	`    |-|||||||||||||o||-||`
	`3'- GUCUUCUCUAAGACCUACUUU-5'`
		latent

y6; gds3; gds18	`5'- CUGAAGAGAUUCUGGGUGUAA-3'`
	`    |-||||-||||||||o|||||`
	`3'- GUCUUCACUAAGACCUACAUU-5'`
		green mosaic

ls11; ls8	`5'- CUGAAGAGAUUCUGGGUGUAA-3'`
	`    |-||||-||||||||o||-||`
	`3'- GUCUUCACUAAGACCUAC UU-5'`
		latent

ls11 (+283U;U271A)	`5'- CUGAAGAGAUUCUGGGUGUAA-3'`
	`    |-|||||||||||||o|||||`
	`3'- GUCUUCUCUAAGACCUACAUU-5'`
		peach yellow mosaic

ls1	`5'- CUGAAGAGAUUCUGGGUGUAA-3'`
	`    |-||||-||||||||o-o-||`
	`3'- GUCUUCACUAAGACCUUU UU-5'`
		latent

ls 14b; esc10; esc14	`5'- CUGAAGAGAUUCUGGGUGUAA-3'`
	`    |-||||-||||||||o-|-||`
	`3'- GUCUUCACUAAGACCUUC UU-5'`
		latent

v1.1	`5'- CUGAAGAGAUUCUGGGUGUAA-3'`
	`    |-||-|-||||||||o||-||`
	`3'- GUCUCCACUGAGACCUAC UU-5'`
		latent

[a] On each duplex, cpSecA mRNA is on top (5´→3´ orientation) and PLMVd-sRNAs below (3´→5´orientation). Position A283 (U283 in the plus polarity) in PYM-sRNA40 is in red and changes in the corresponding sRNAs from other variants are in blue. |, Watson-Crick base pairs; o, G:U wobble base pairs; -, mismatches or bulges