Supplementary Figures: Figure S1~S31

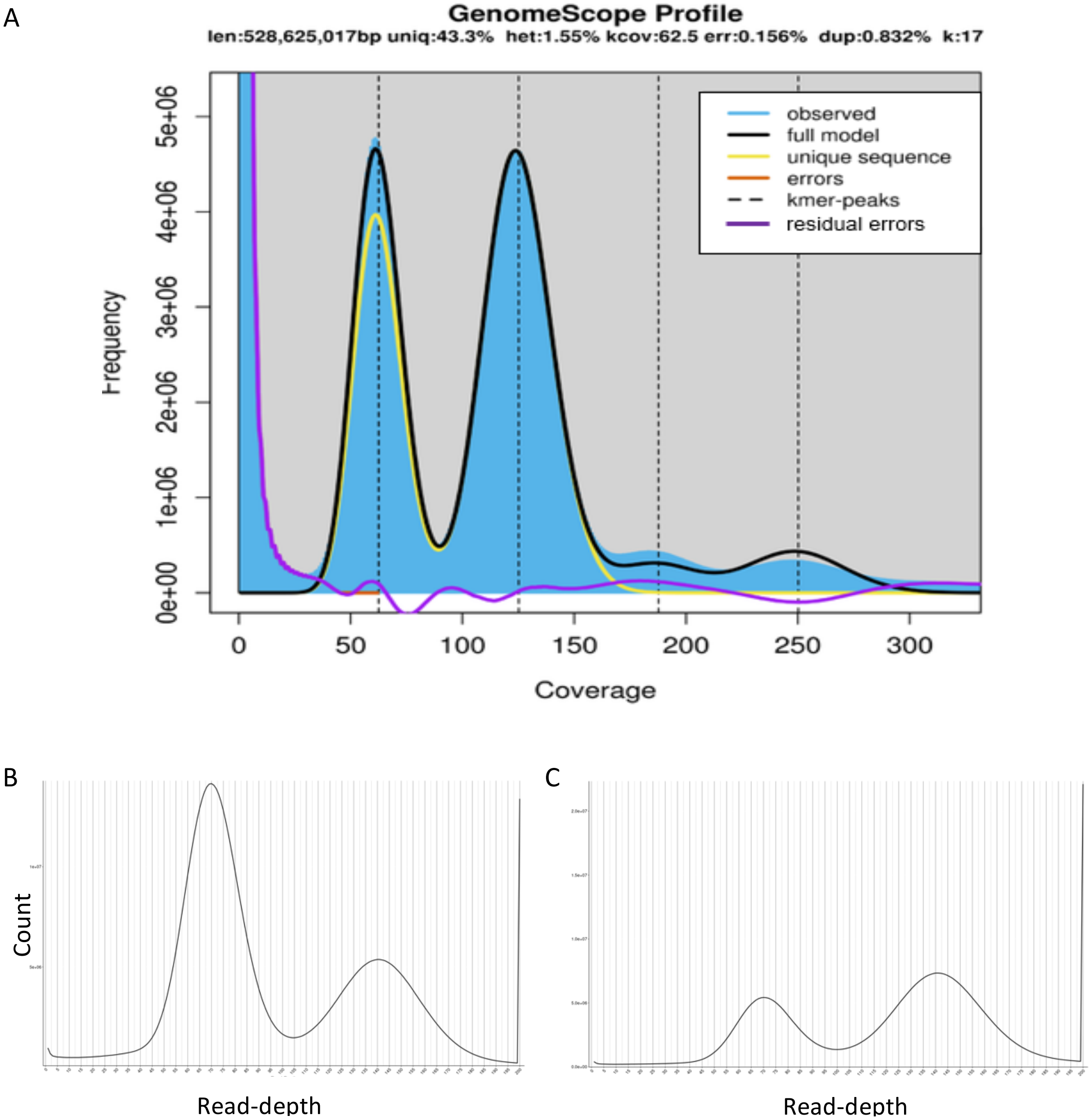


Figure S1 Evaluation of *Rhododendron ovatum* genome by *k*-mer analysis and read-depth. A. *k*-mer analysis. B. Read-depth before elimination of redundancy. C. Read-depth after elimination of redundancy.

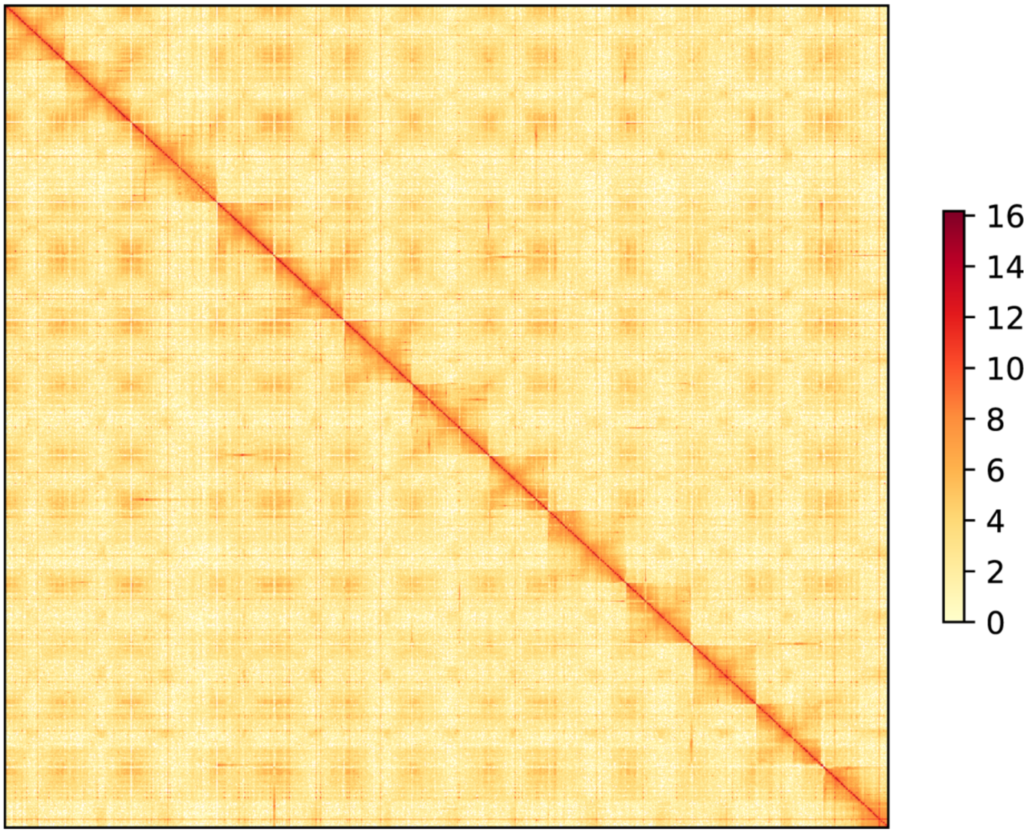


Figure S2 Genome-wide analysis of chromatin interactions in the *R. ovatum* genome based on Hi-C data. The resolution is 500 Kb.

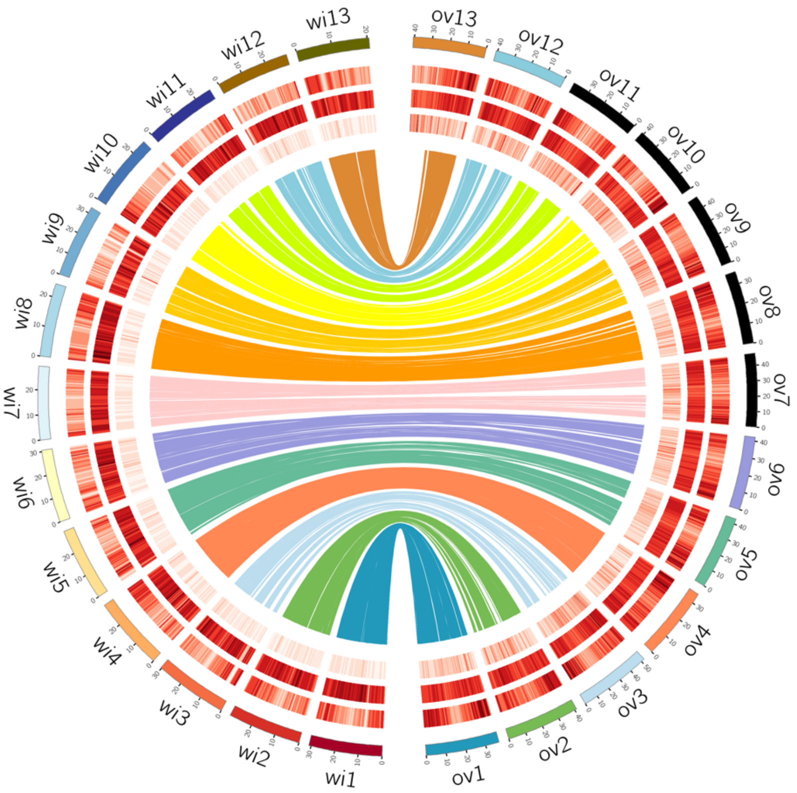


Figure S3 Genome assemblies of *R. ovatum* (ov) and *R. williamsianum* (wi). From outer to inner is the pseudochromosomes, gene density, transposon elements, tandem repeats and collinear blocks of the two genomes.

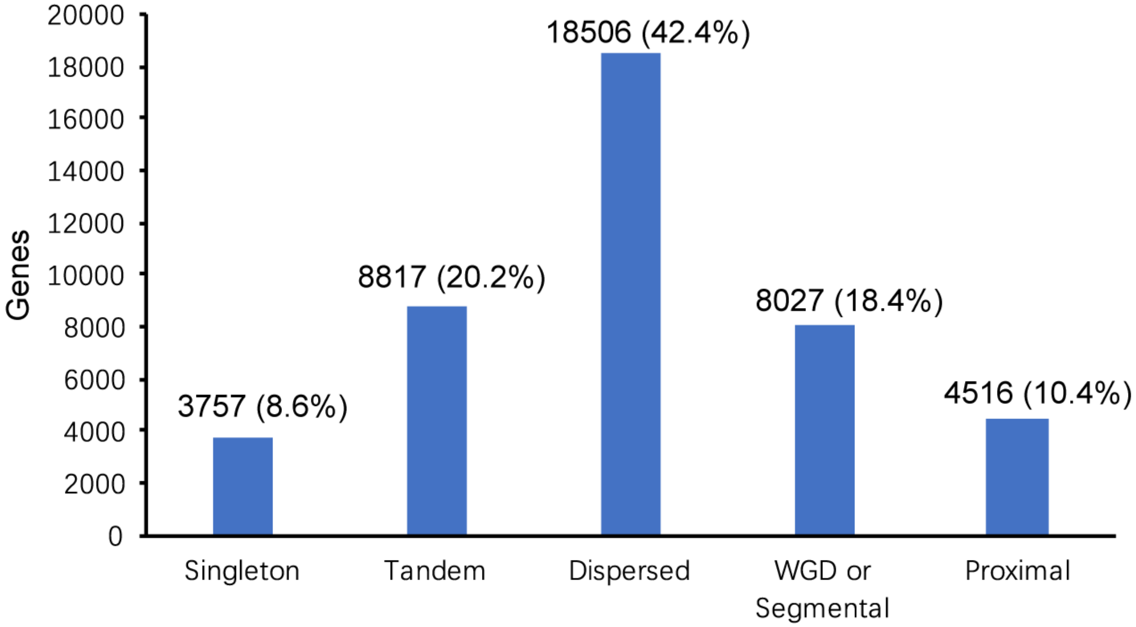


Figure S4 Types of gene duplication in the *R. ovatum* genome. Singleton means that the gene is single-copy, which should not be the type of members of gene families. Tandem means multiple members of one family occurring within the same intergenic region or in neighboring intergenic regions. Dispersed means that the gene might arise from transposition, such as 'replicative transposition', 'non-replicative tranposition' or 'conservative tranpostion'. Proximal means that the gene might arise from small-scale transposition or arise from tandem duplication and insertion of some other genes. WGD or segmental means that the gene might arise from Whole Genome Duplication or Segmental Duplication.

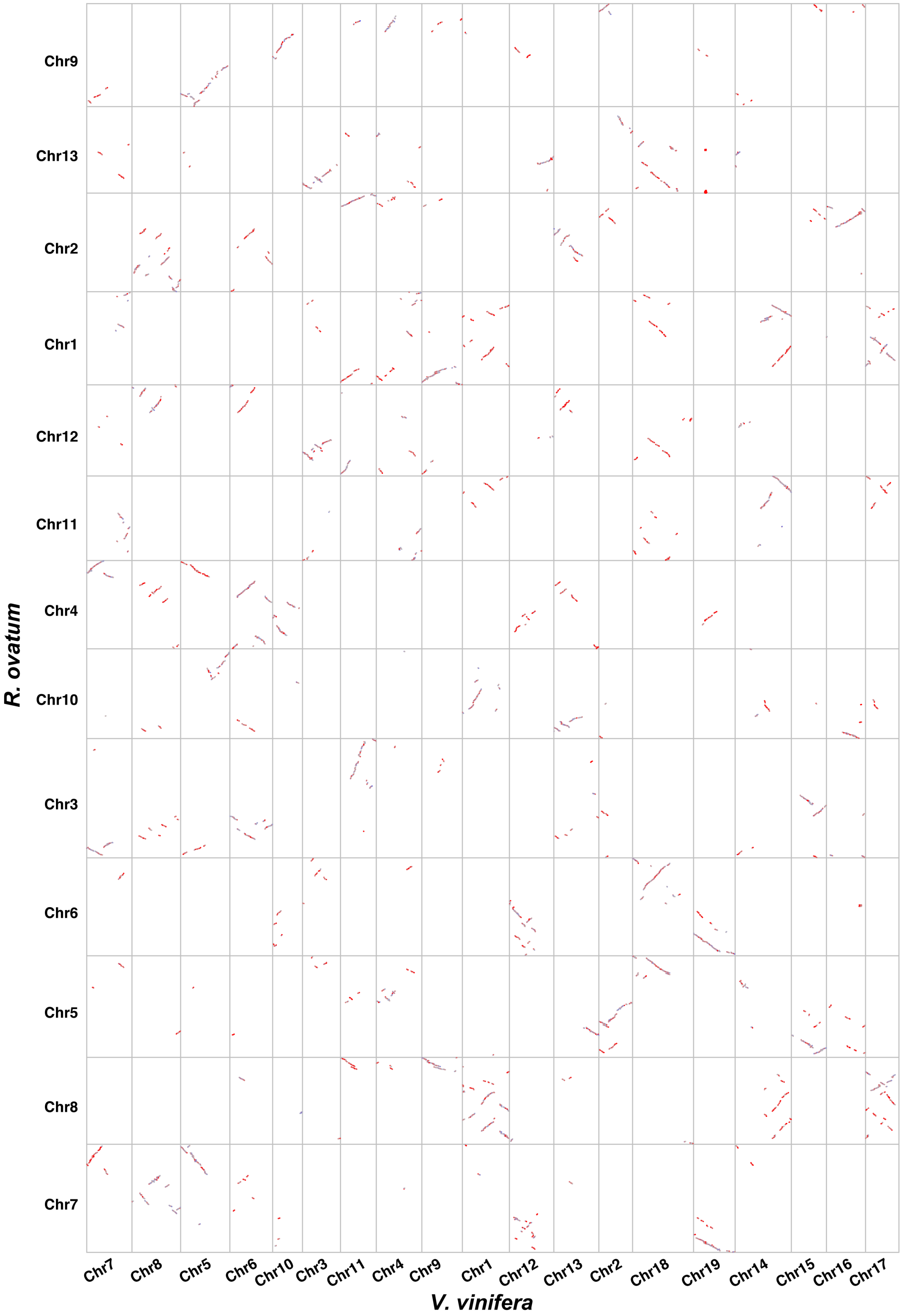


Figure S5 ***Ks*** **dot plot between** ***R. ovatum*** **and grape (*****V. vinifera*****).**

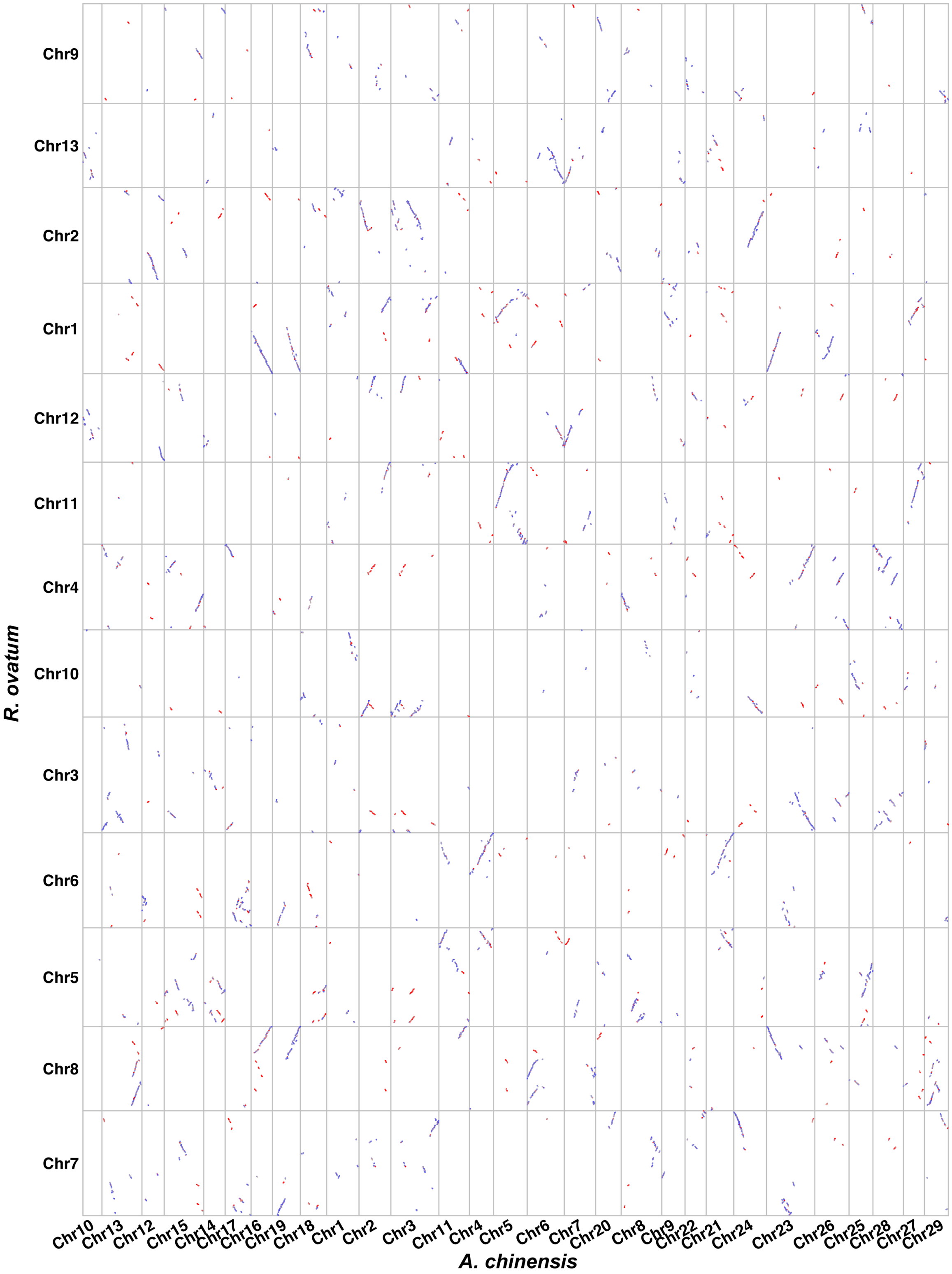


Figure S6 ***Ks*** **dot plot between** ***R. ovatum*** **and kiwifruit (*****A. chinensis*****).**

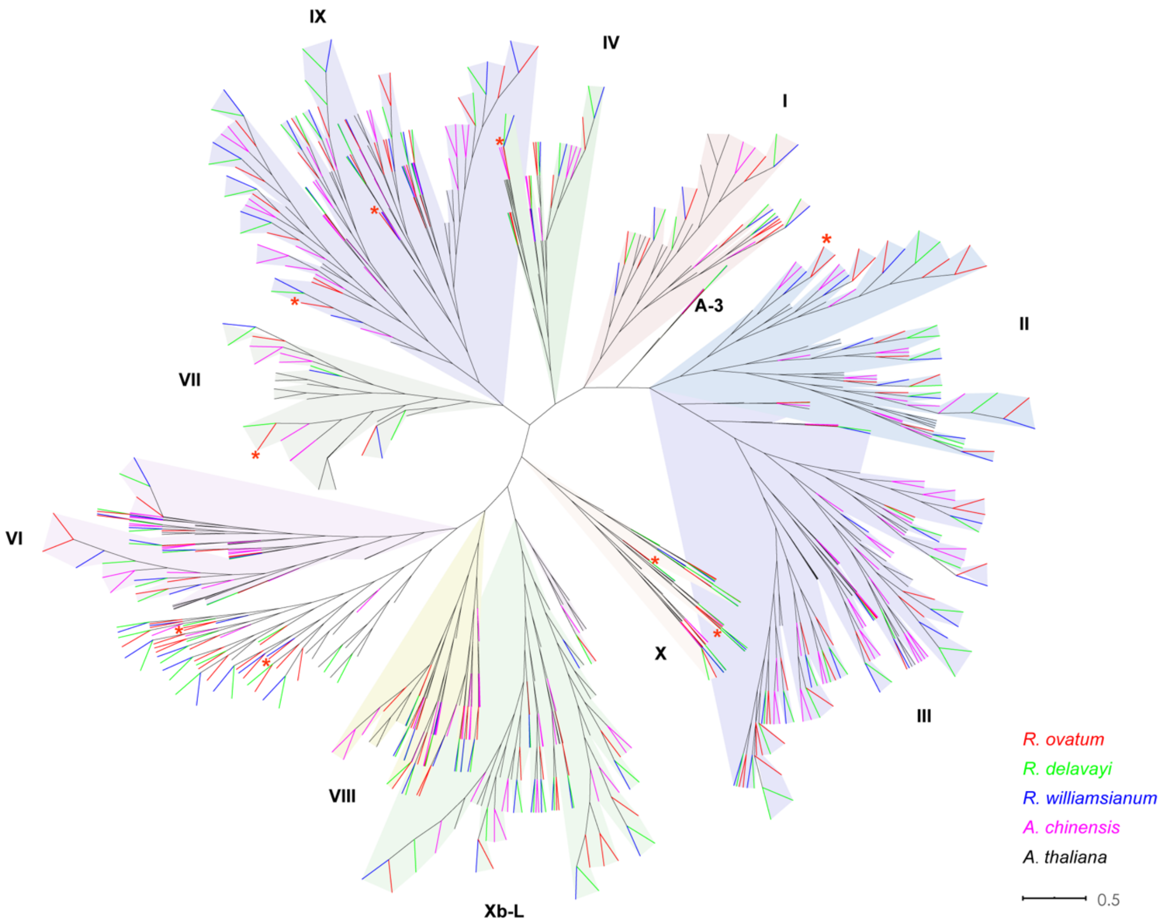


Figure S7 Phylogenetic tree of *ERF* gene family. Branches are color-coded according to species. The characters beside each clade represent the subfamily names. Asterisks (*) indicate the genes that have undergone positive selection (Ka/Ks>1).

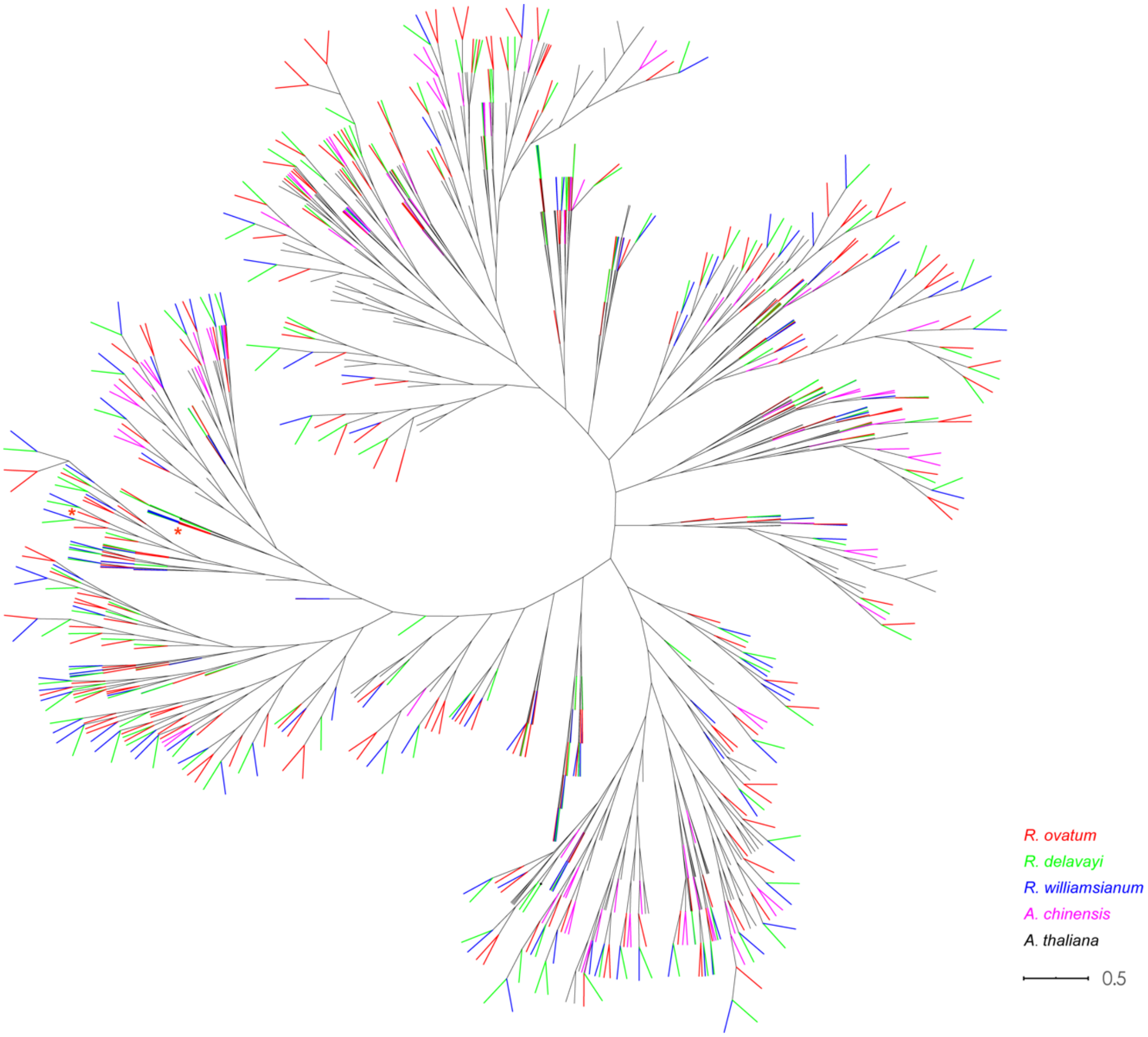


Figure S8 Phylogenetic tree of *MYB* gene family. Branches are color-coded according to species. Asterisks (*) indicate the genes that have undergone positive selection (Ka/Ks>1).

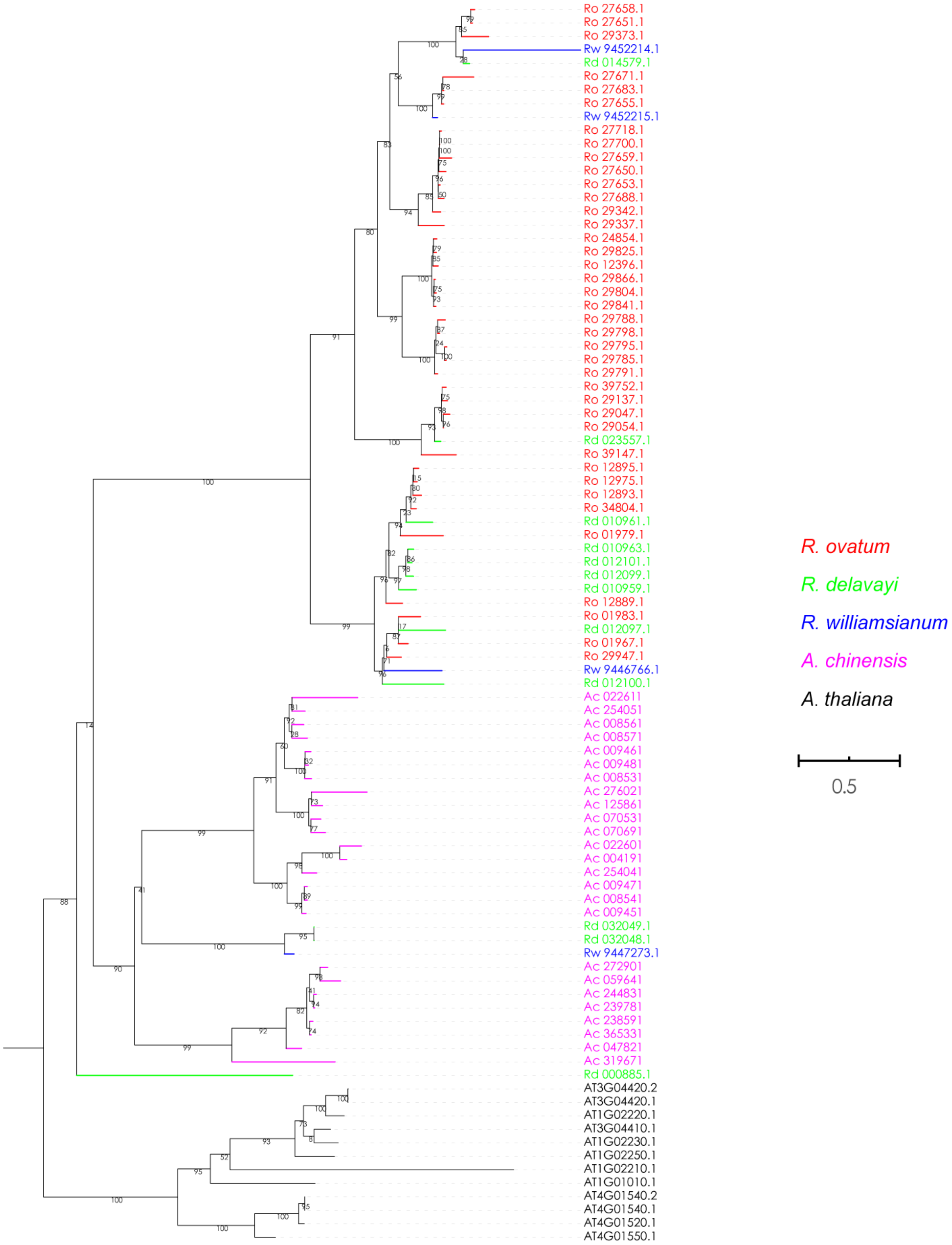


Figure S9 Phylogenetic tree of *ANAC001* clade of *NAC* gene family. The gene number of the ANAC001 clade in *R. ovatum* was significantly expanded, which is much larger than that of other species.

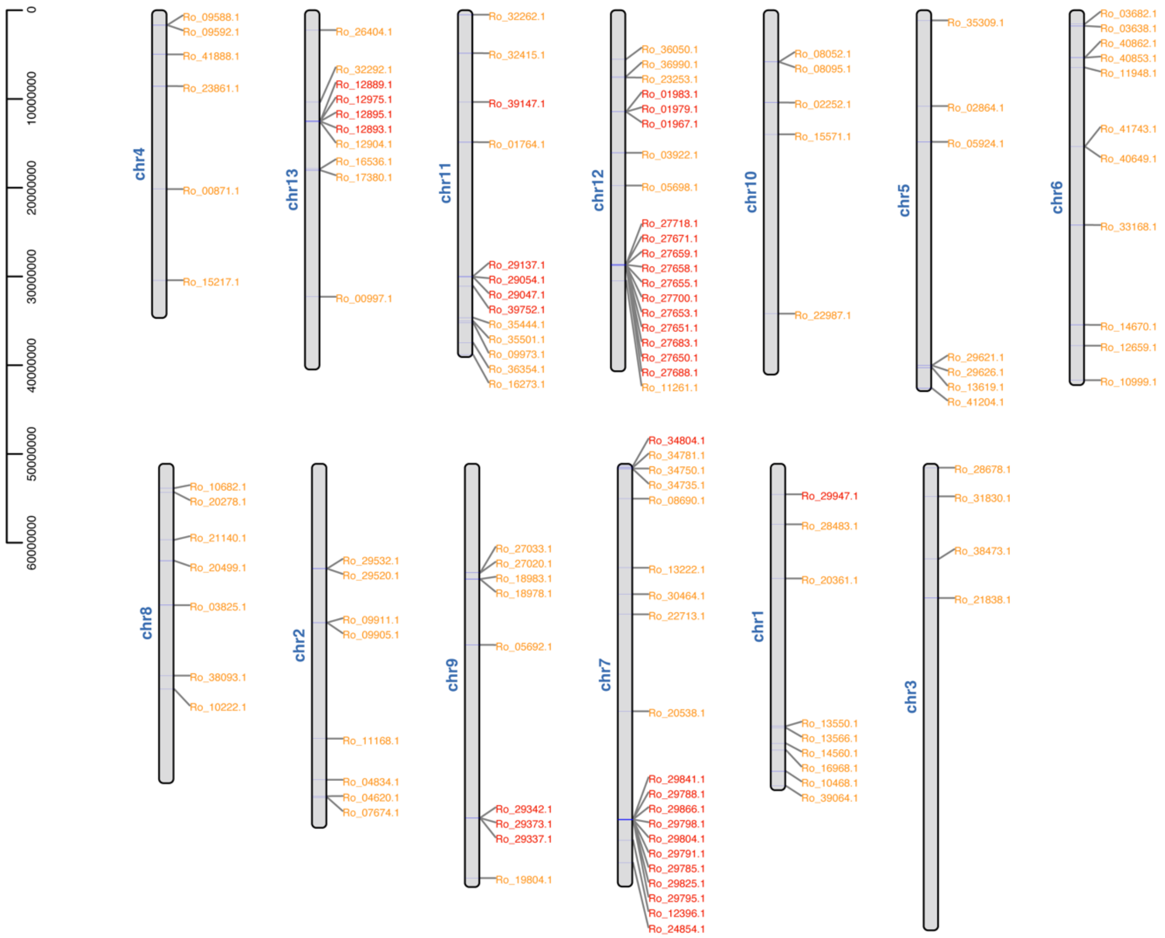


Figure S10 Chromosome localization of *NAC* genes of *R. ovatum*. The genes in red indicate members of ANAC001 clade, which show several tandem repeat clusters.

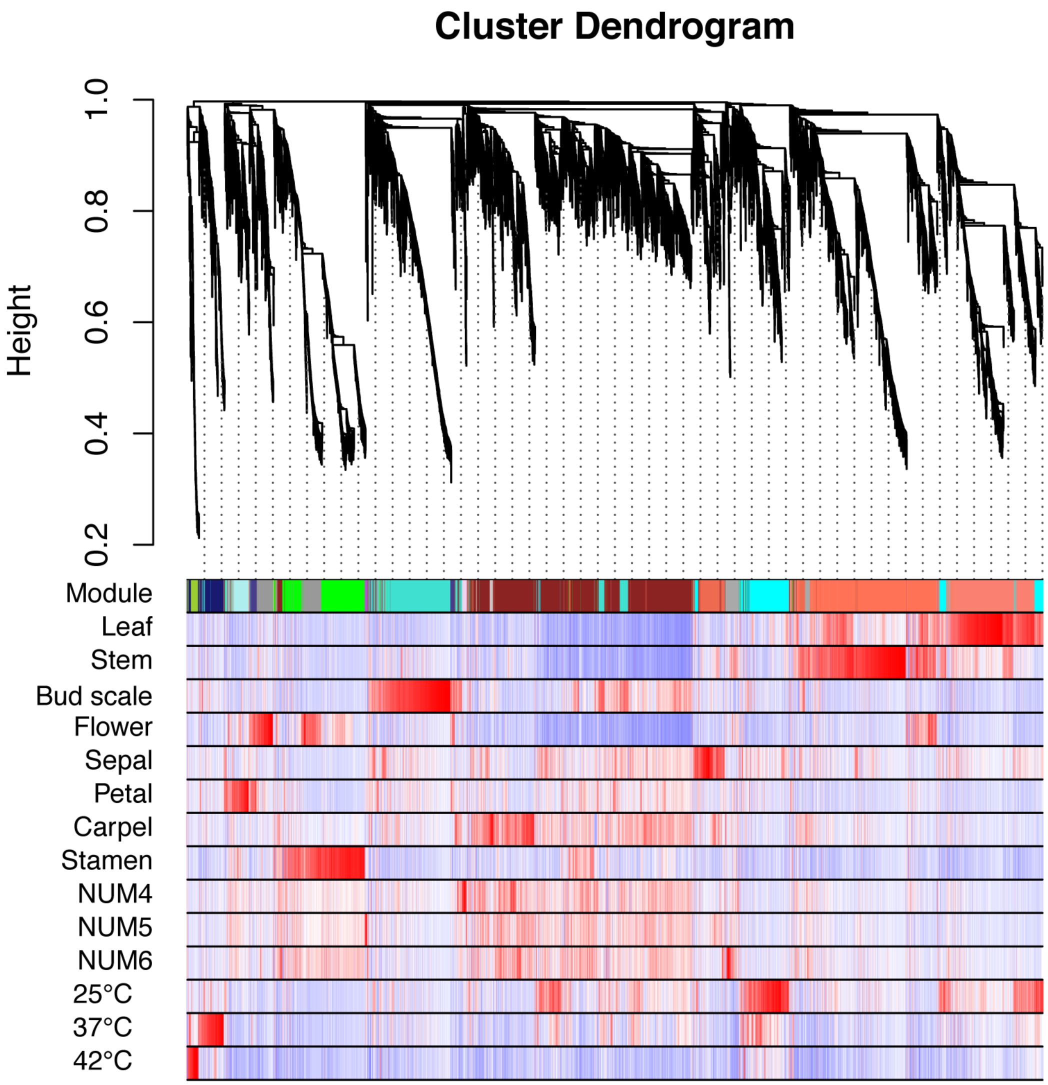


Figure S11 Weighted gene co-expression network analysis (WGCNA) of the transcriptomes. Modules of same color represent the genes that have similar expression profiles.

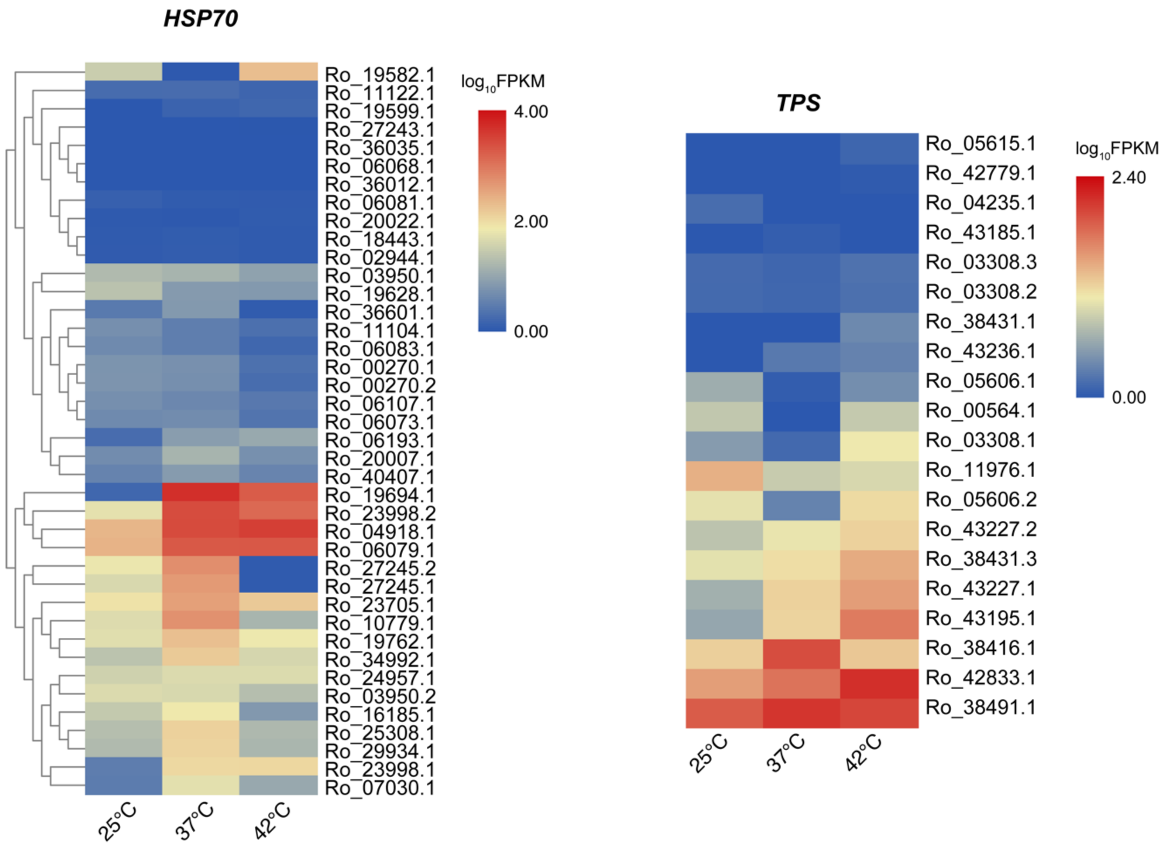


Figure S12 Expression profiles of *HSP70s* and *TPSs* in response to different temperature treatments (25, 37 and 42℃).

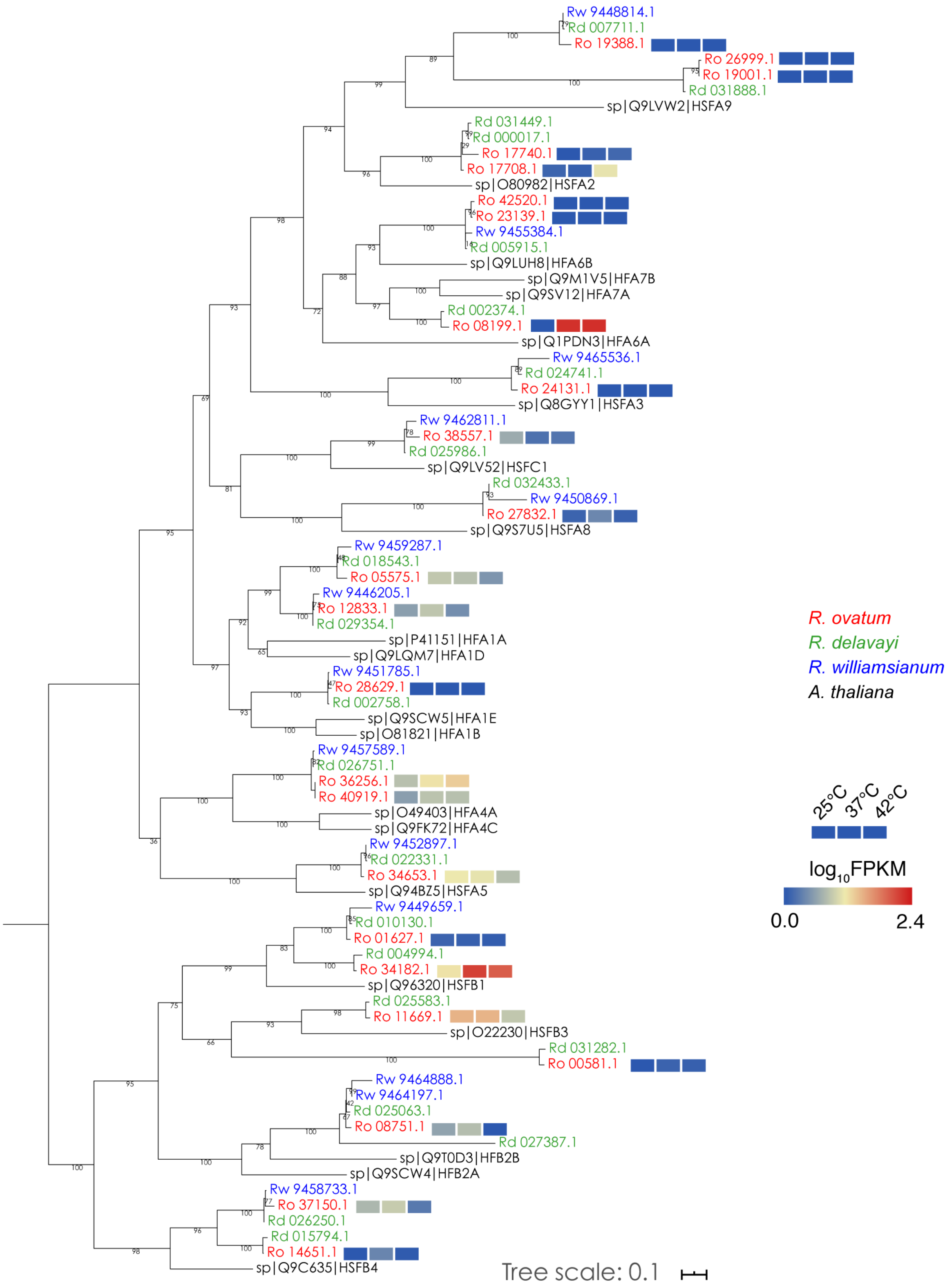


Figure S13 Phylogenetic analysis and heat-induced expression profiles of *HSFs* in *R. ovatum*.

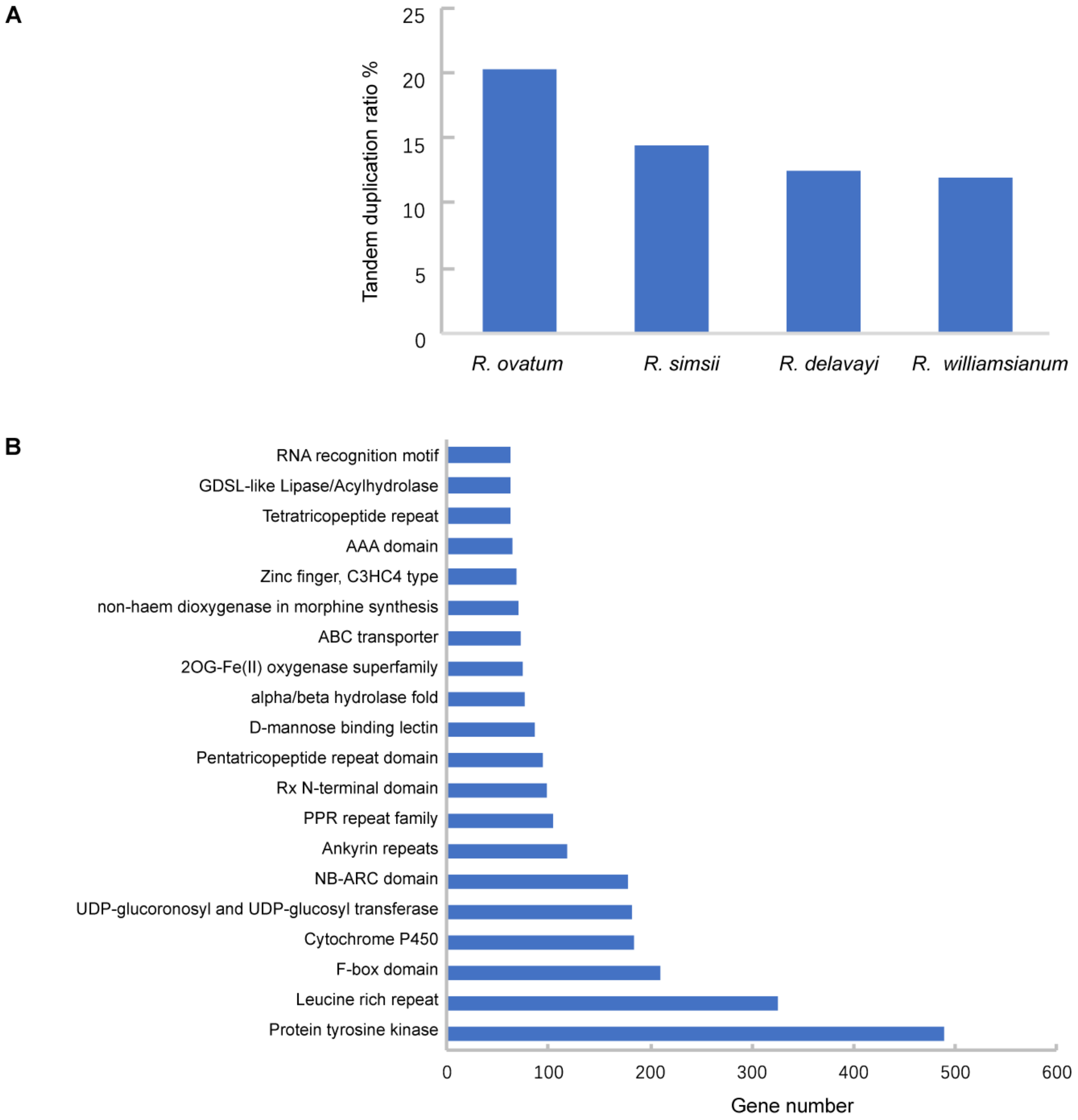


Figure S14 Characteristics of tandem duplication in *R. ovatum* genome. A. Tandem duplication ratio of *R. ovatum* compared with the other three *Rhododendron* species. B. The top 20 Pfam domains contained in tandem-duplicated genes of *R. ovatum*.

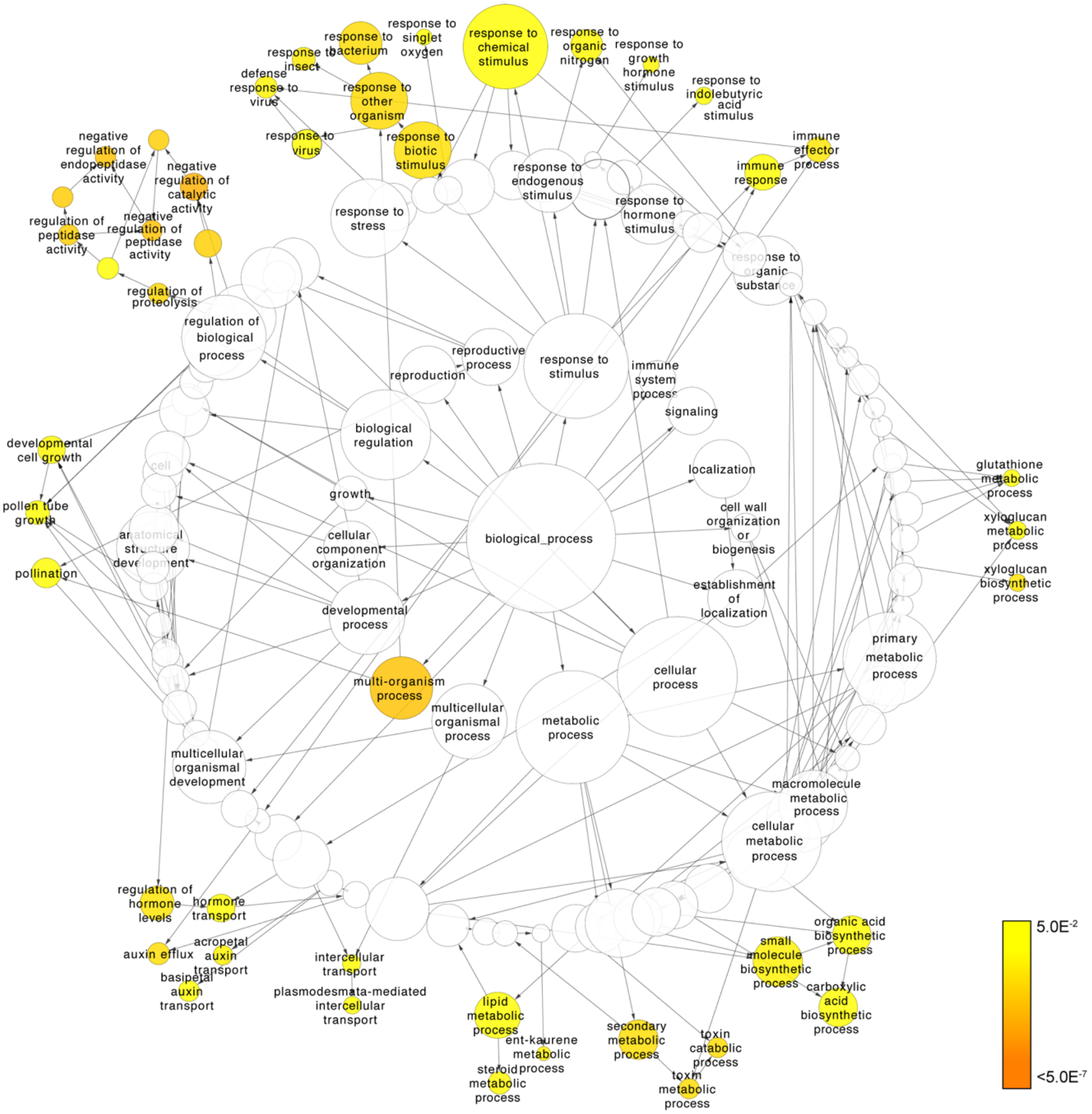


Figure S15 GO functional enrichment network of tandem-duplicated genes. All the genes generated by tandem duplication were performed GO annotation and enrichment. The relationships of GO terms were visualized using BiNGO in Cytoscape. Size of circles indicates gene numbers included. Circles in yellow or orange color indicate the GO terms that have significant enrichment match (adjusted p-value <0.05).

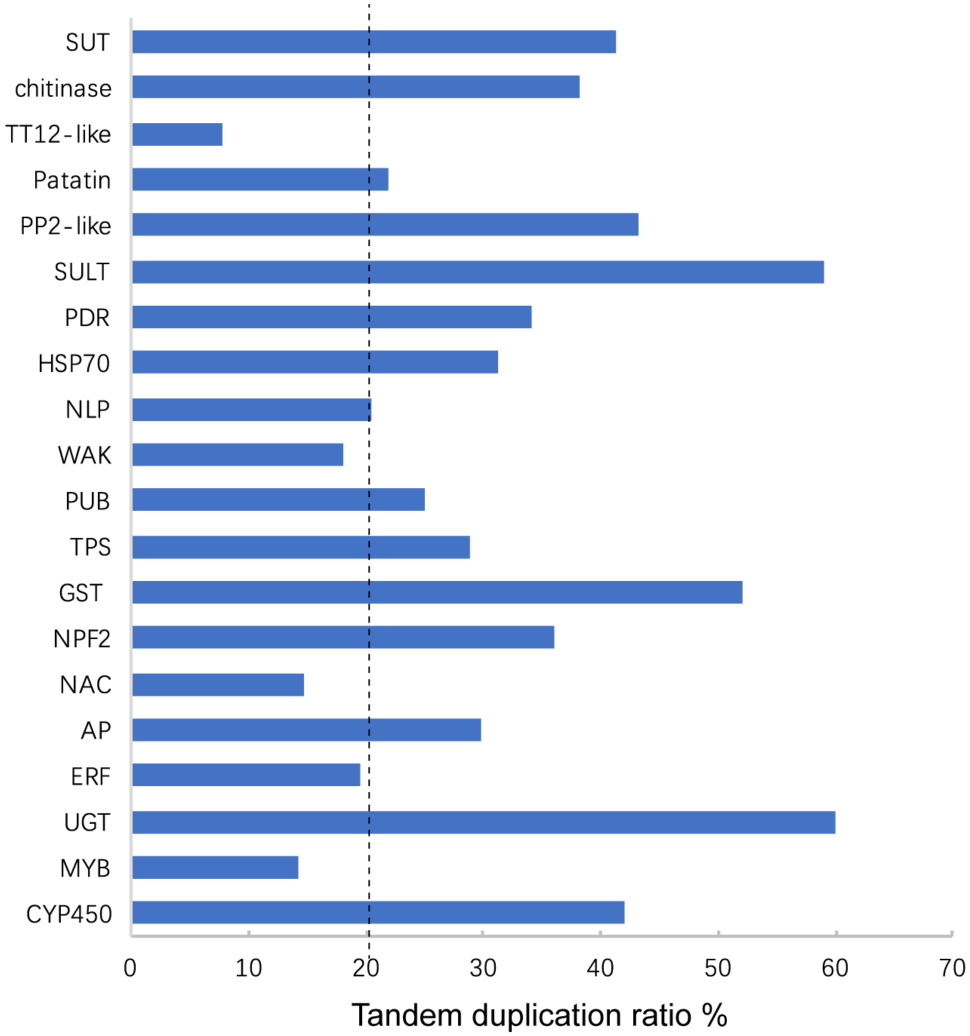


Figure S16 Tandem duplication ratio of the stress responsive genes. The tandem duplication ratio was calculated as: family genes generated by tandem duplication / total genes of the family. The dotted line indicates the average tandem duplication ratio (20.21%) of the whole genome of *R. ovatum*. The 15 of 20 gene families have higher tandem duplication ratios than the average level of the whole genome.

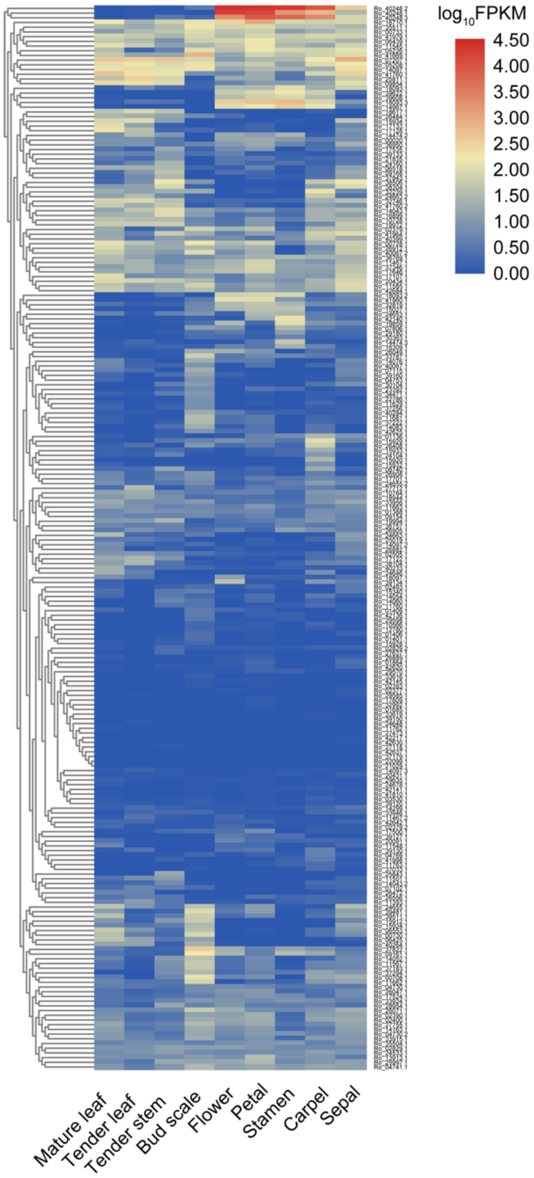


Figure S17 Expression profiles of CYP450s in different tissues. Three alternative splicing transcripts of one *CYP450* (Ro_40248) have highest transcriptional levels in floral tissues.

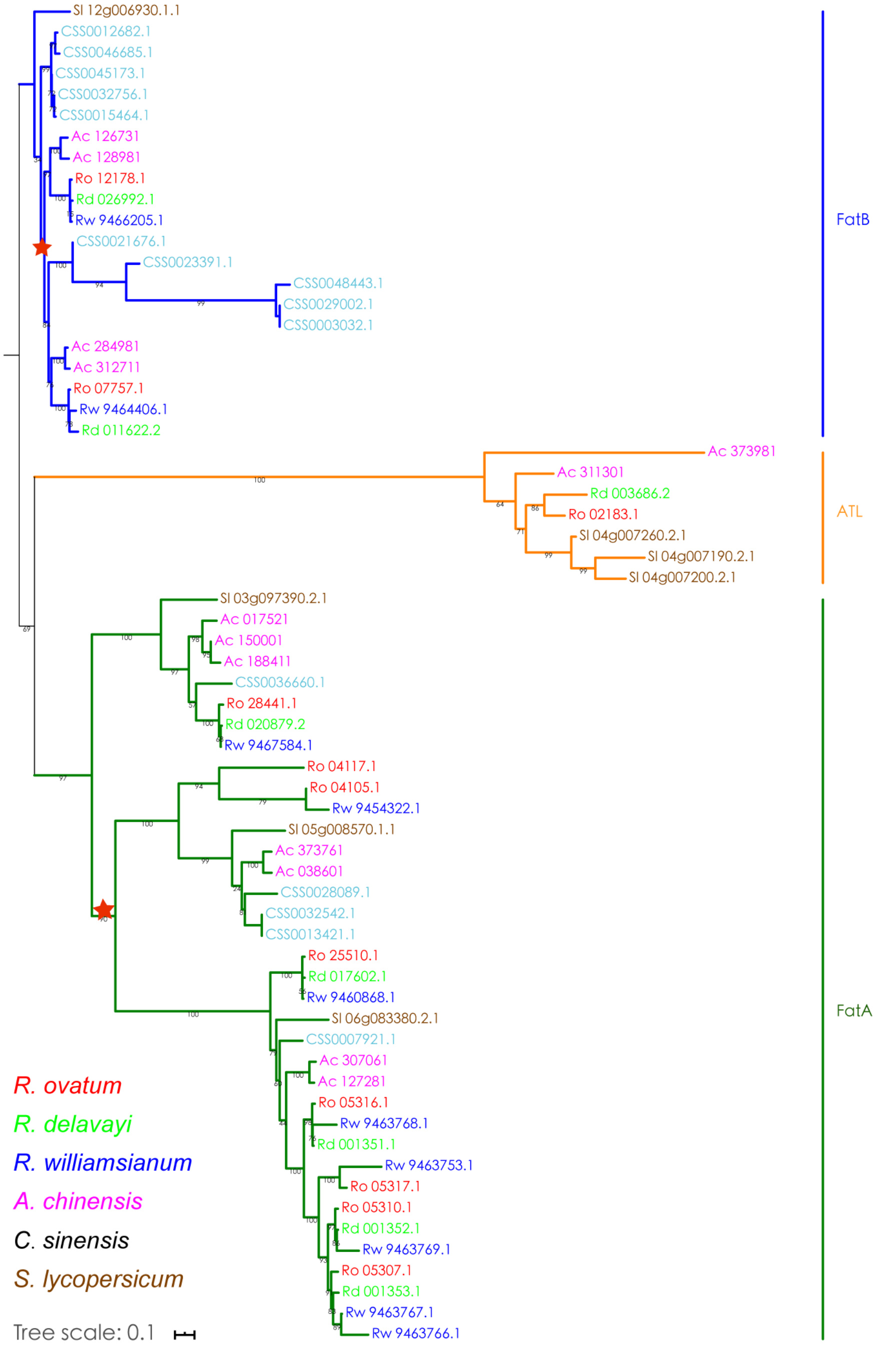
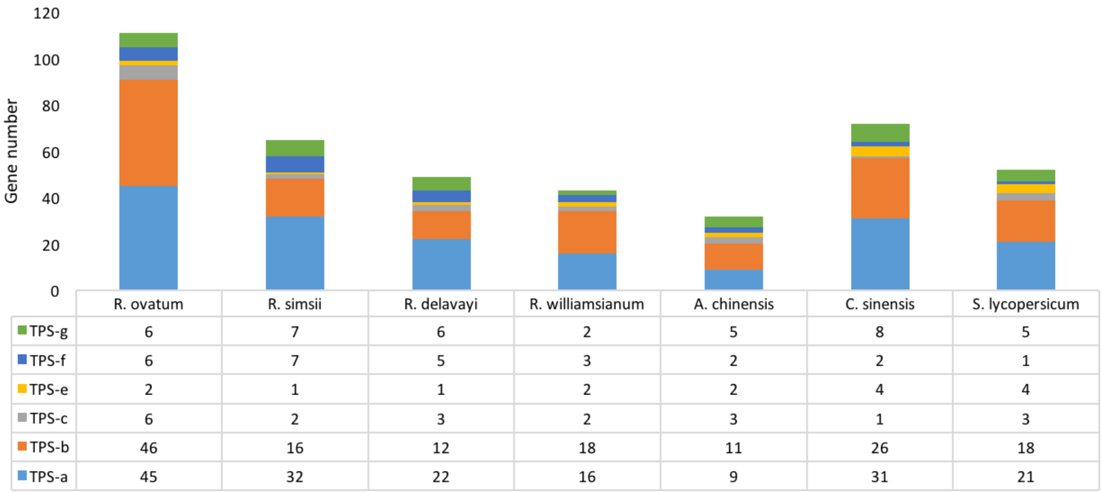


Figure S18 Phylogenetic tree of fatty acyl-ACP thioesterases coding genes. The asterisks indicate WGD events.



	R. ovatum	R. simsii	R. delavayi	R. williamsianum	A. chinensis	C. sinensis	S. lycopersicum
🟩 TPS-g	6	7	6	2	5	8	5
🟦 TPS-f	6	7	5	3	2	2	1
🟨 TPS-e	2	1	1	2	2	4	4
⬜ TPS-c	6	2	3	2	3	1	3
🟧 TPS-b	46	16	12	18	11	26	18
🟦 TPS-a	45	32	22	16	9	31	21

Figure S19 Statistic of *TPS* gene subfamilies. *R. ovatum* has much more *TPS* genes than other species. The *RoTPS* expansion mainly occurred in subfamily b.

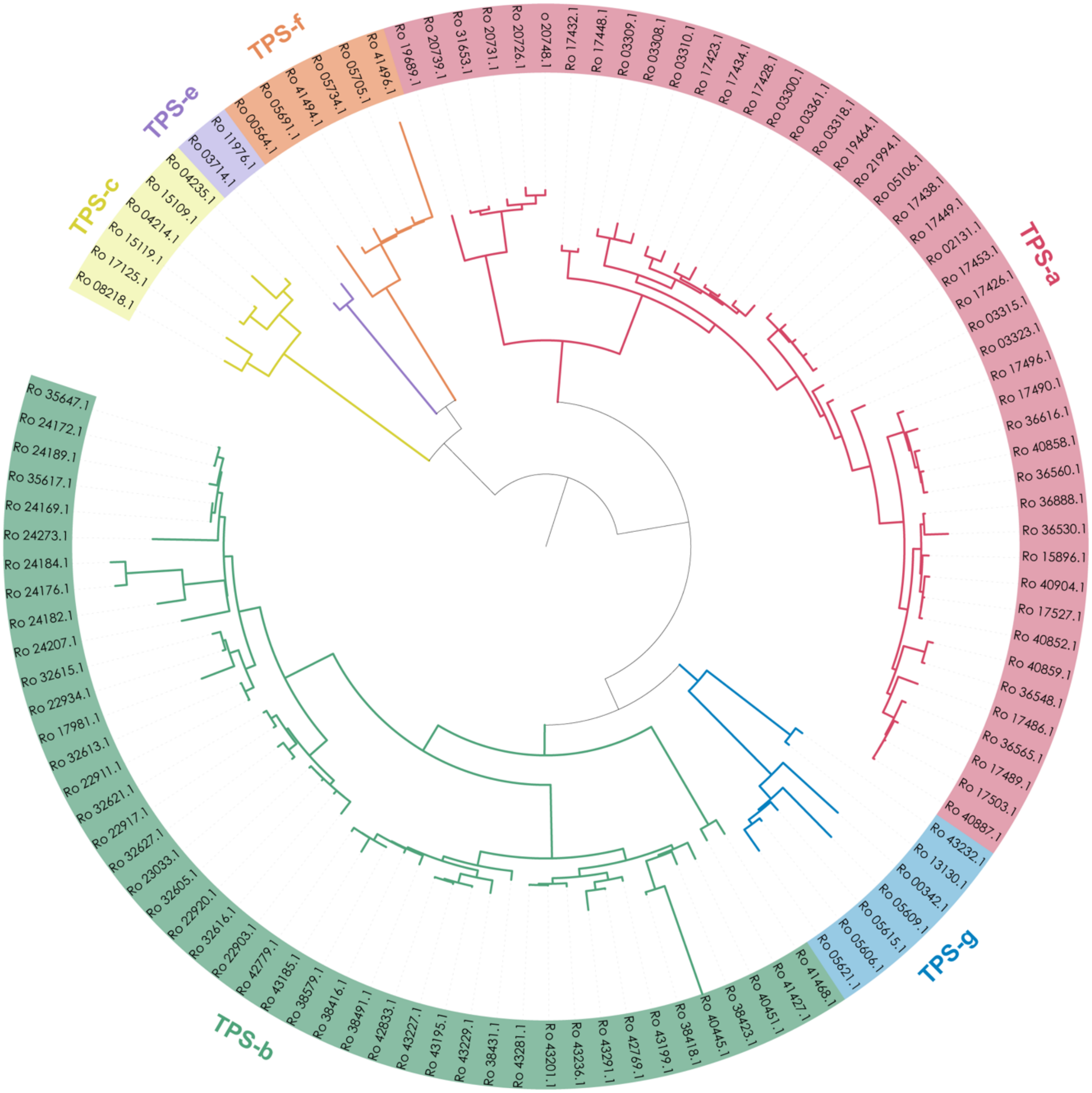


Figure S20 Phylogenetic tree of TPSs in *R. ovatum*.

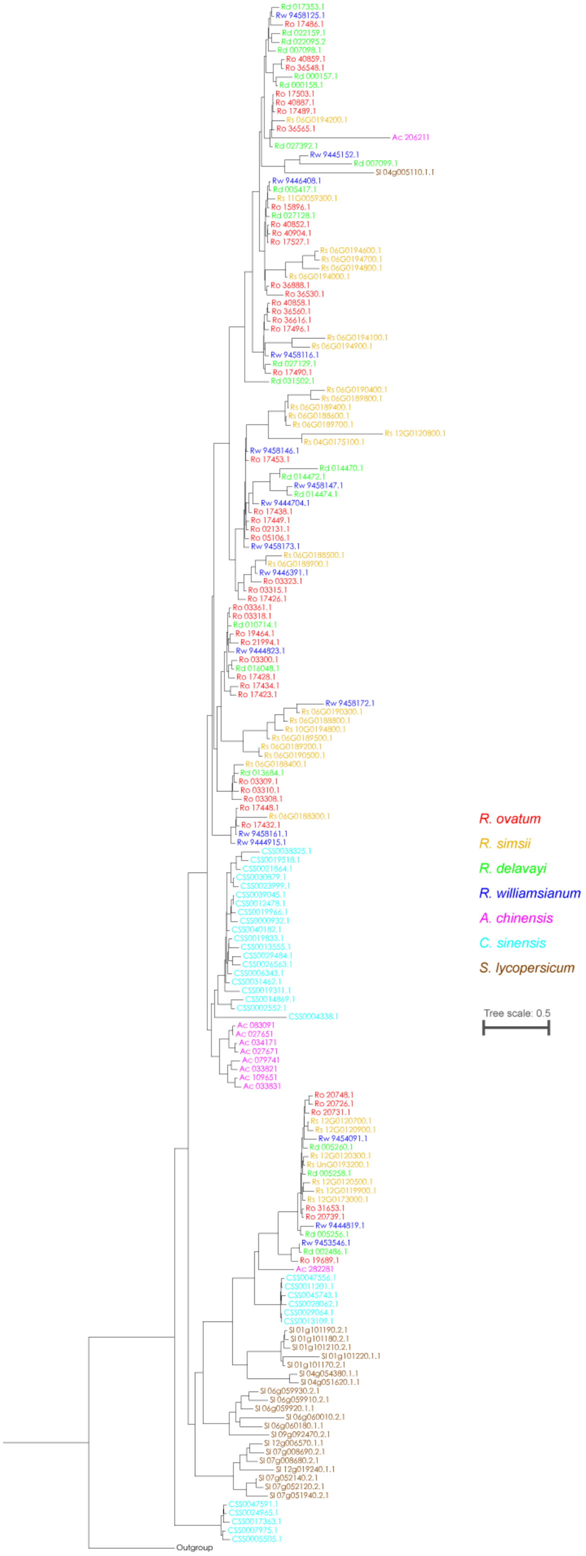


Figure S21 Phylogeny of the TPS-a subfamily. A tomato TPS of TPS-g is chosen as the outgroup. RoTPS-a has occurred duplications for most members, thereby significantly expanded the subfamily.

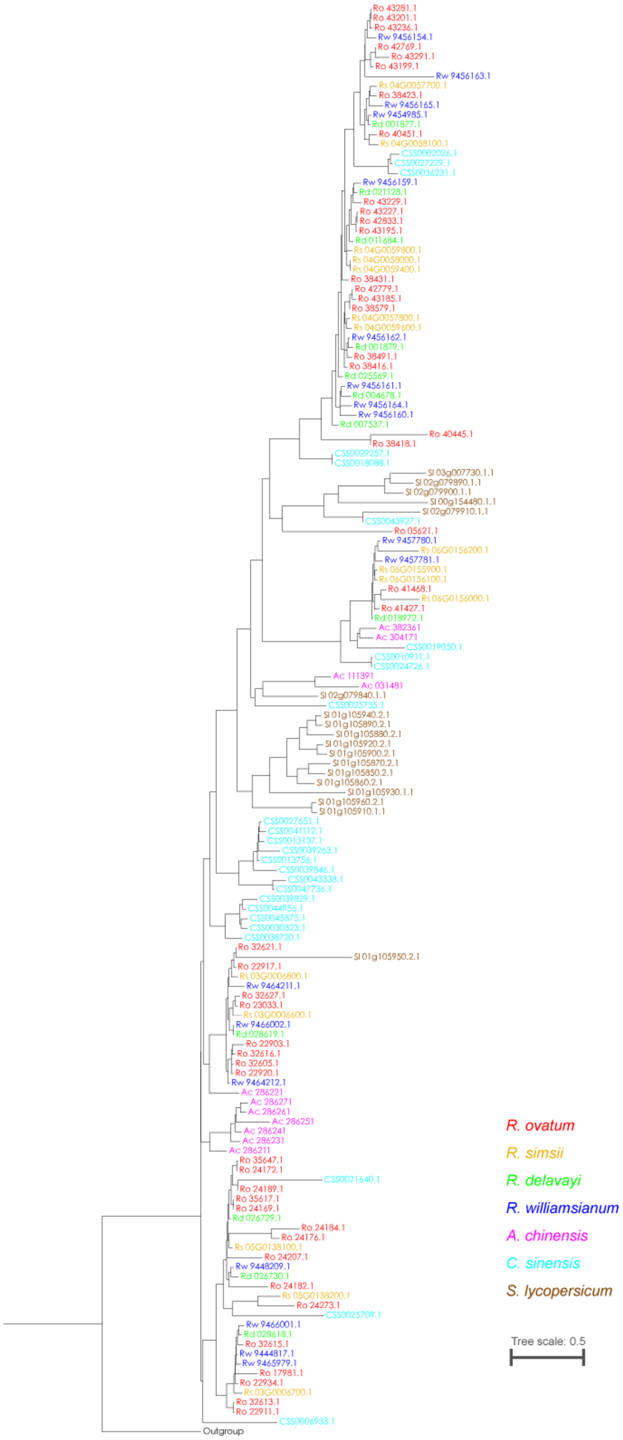


Figure S22 Phylogeny of the TPS-b subfamily. A tomato TPS of TPS-g is chosen as the outgroup. The TPS-b subfamily of *R. ovatum* also has duplicated several times to generate larger group.

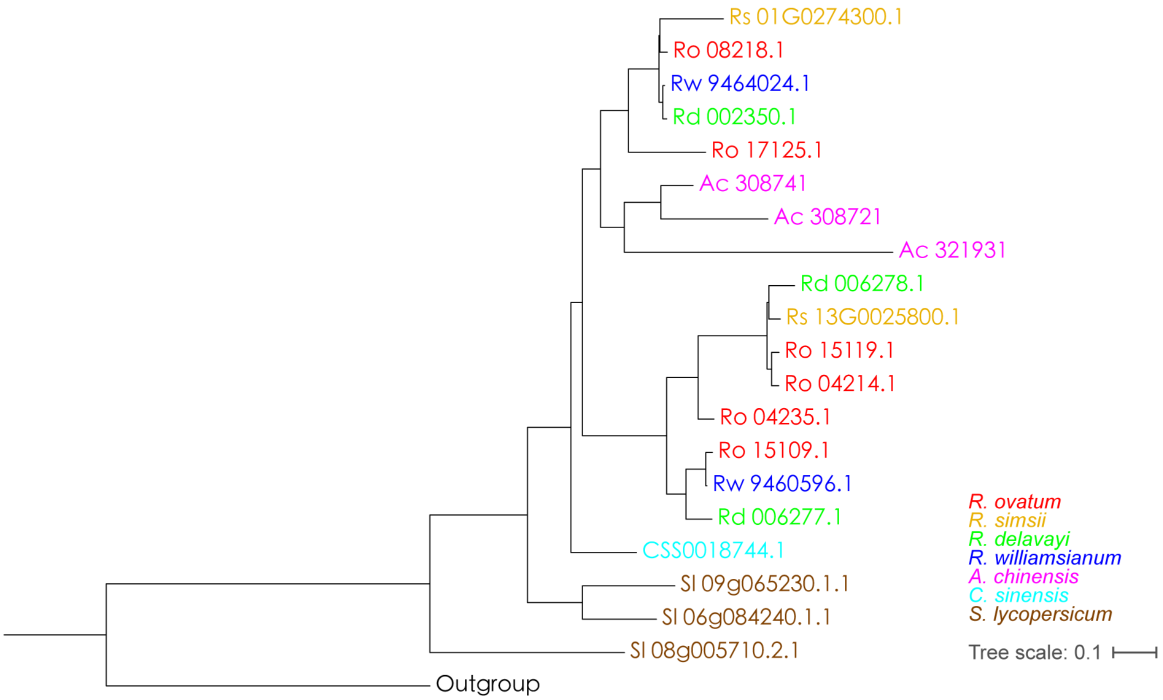


Figure S23 Phylogeny of the TPS-c subfamily. A tomato TPS of TPS-e is chosen as the outgroup. Although TPS-c is a small subfamily, number of RoTPS-c is still at least two folds of that in other species.

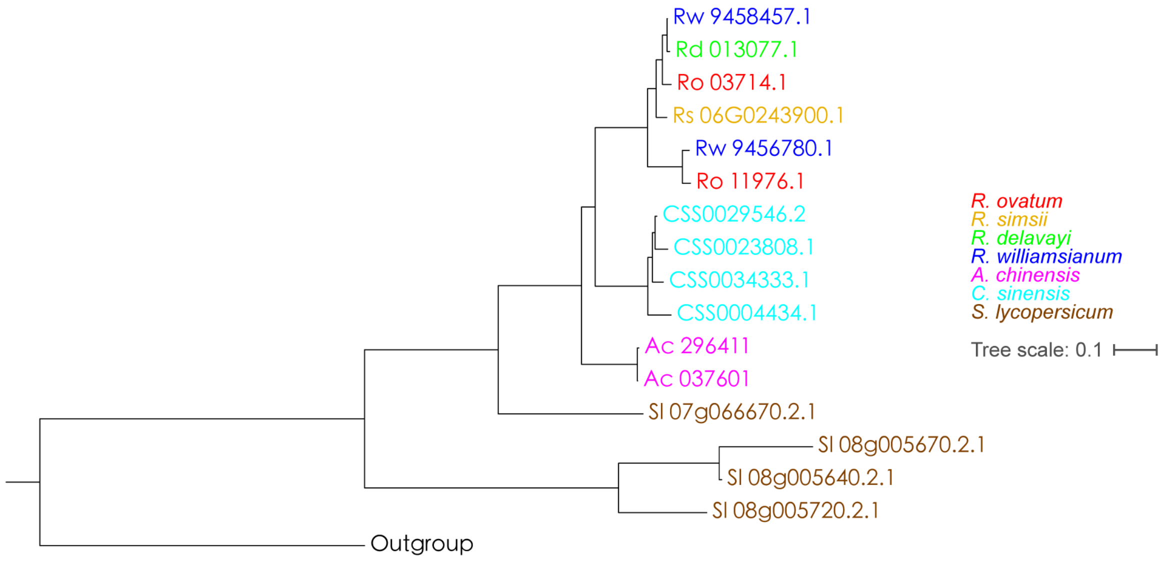


Figure S24 Phylogeny of the TPS-e subfamily. A tomato TPS of TPS-f is chosen as the outgroup. RoTPS-e has not expanded.

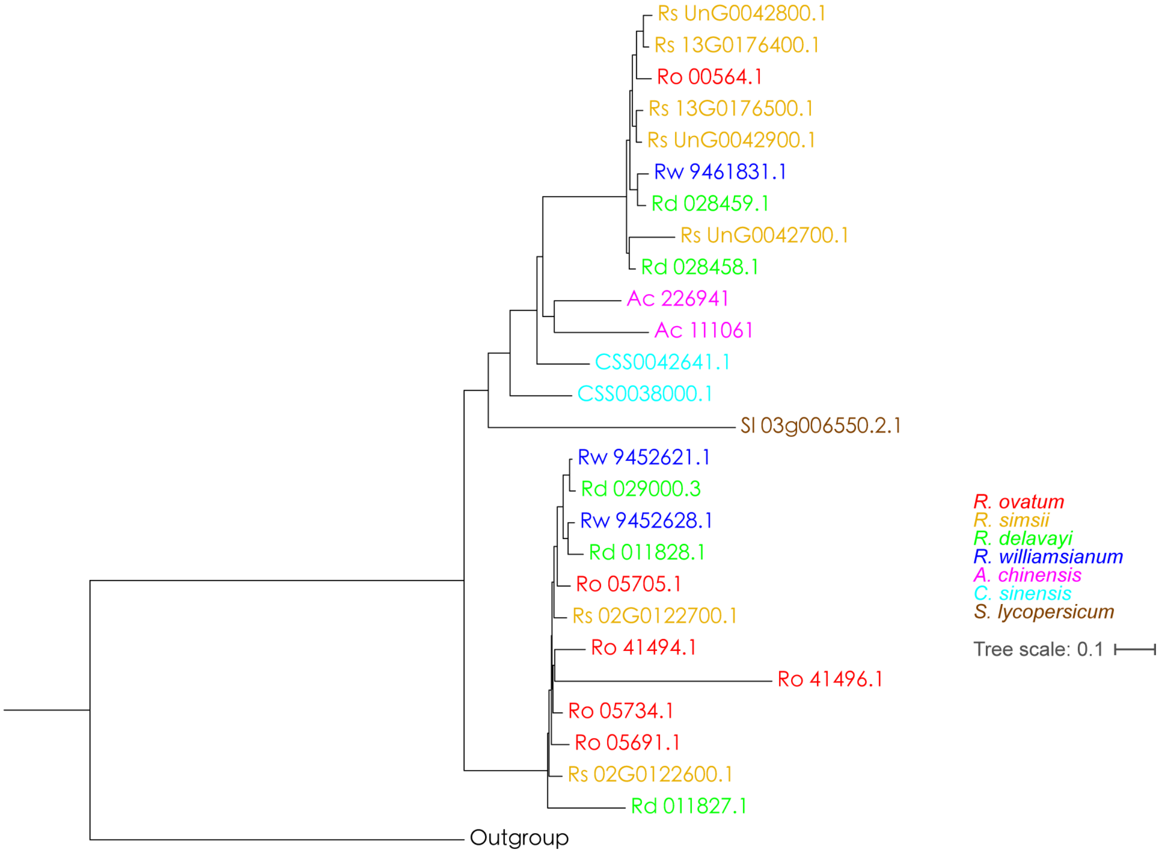


Figure S25 Phylogeny of the TPS-f subfamily. A tomato TPS of TPS-e is chosen as the outgroup. *Rhododendron* TPS-f has expanded compared to kiwi fruit, tea and tomato, but RoTPS-f has not significant expansion than the other three rhododendrons.

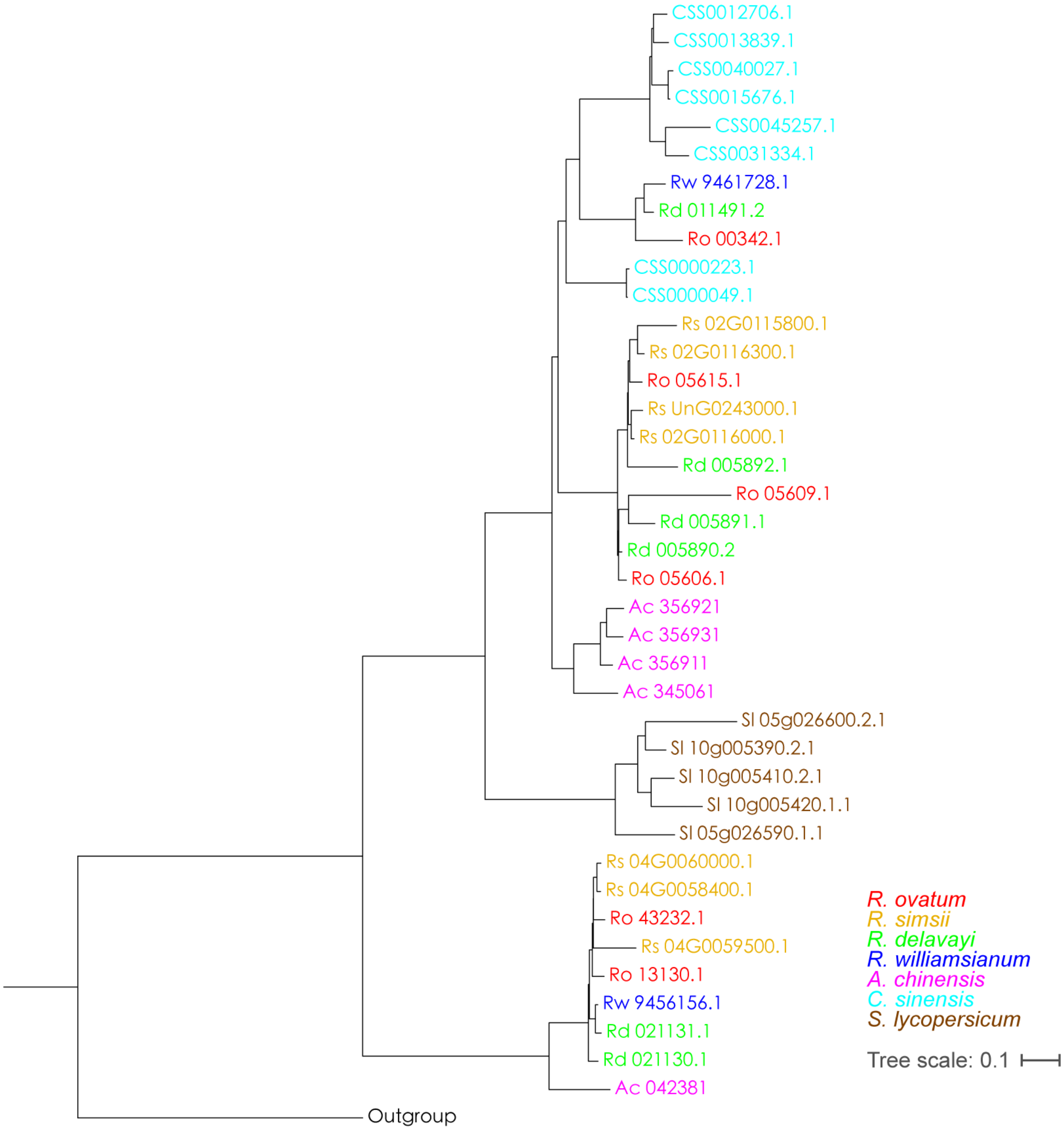


Figure S26 Phylogeny of the TPS-g subfamily. A tomato TPS of TPS-e is chosen as the outgroup. RoTPS-g has not expanded.

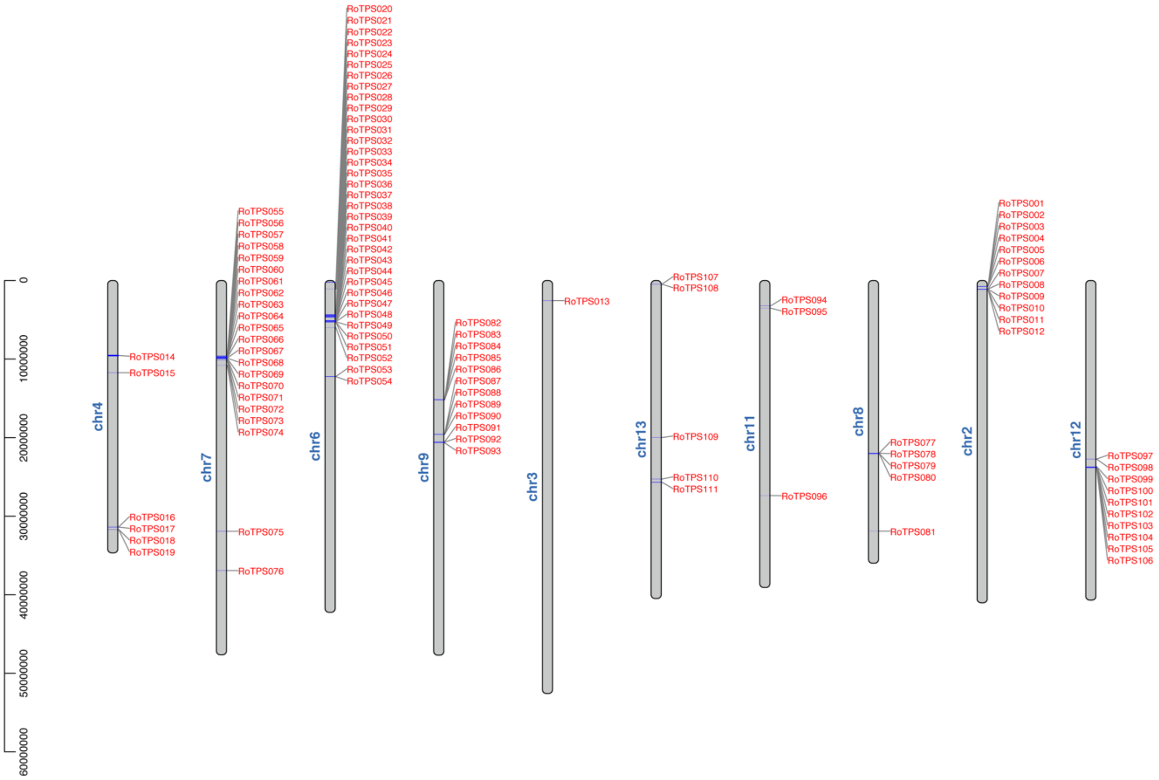


Figure S27 Chromosome localization of *RoTPS* genes. *TPSs* are distributed on 10 of 13 chromosomes of *R. ovatum*, with most tandem density on chr6 and chr7.

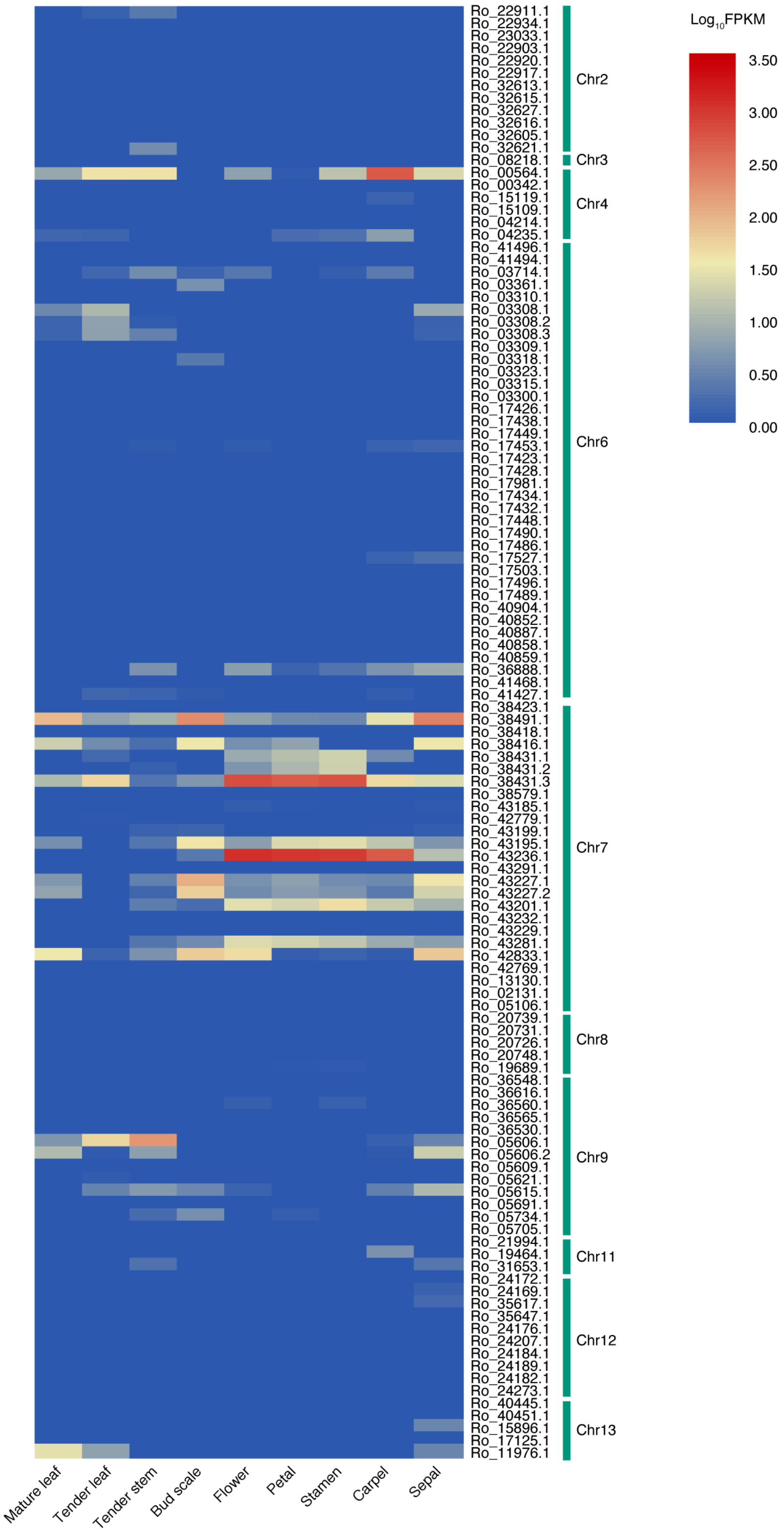


Figure S28 Expression profiles of *TPS* genes of *R. ovatum*. The *TPS* genes that have high expression levels are mainly distributed on chr7.

A

```
Ro_38431.2      MSLHLFFVPIRSIPLTLGLPPHLTRKPVHSNTHTRLYCLQSSKLVDQDSQQTSQRRCANY  60
Ro_38431.1      MSLHLFFVPIRSIPLTLGLPPHLTRKPVHSNTHTRLYCLQSSKLVDQDSQQTSQRRCANY  60
Ro_38431.3      MSLHLFFVPIRSIPLTLGLPPHLTRKPVHSNTHTRLYCLQSSKLVDQDSQQTSQRRCANY  60
                ************************************************************

Ro_38431.2      QPTAWNSDFIKSLKNHNVTYIHTCMY----------VHGCSSQDDWILVKDEIQKQRAEK  110
Ro_38431.1      QPTAWNSDFIKSLKNHNVLRFELQTSQILELTPSDVQSGKLFSLFYRCLIDEIQKQRAEK  120
Ro_38431.3      QPTAWNSDFIKSLKNHNV--------------------------------DEIQKQRAEK  88
                ******************                                **********

Ro_38431.2      LKEDVRAMIGDTYANSLTILELIDDIQRLGLSYHFDKDIKRALEKIILSMNGNNVMNDQK  170
Ro_38431.1      LKEDVRAMIGDTYANSLTILELIDDIQRLGLSYHFDKDIKRALEKIILSMNGNNVMNDQK  180
Ro_38431.3      LKEDVRAMIGDTYANSLTILELIDDIQRLGLSYHFDKDIKRALEKIILSMNGNNVMNDQK  148
                ************************************************************

Ro_38431.2      IGVHAIALCFRLCRQHGYEVSQDVFKRFKDENGNFMESLSKDTKGLLSLYEASYFSFDGE  230
Ro_38431.1      IGVHAIALCFRLCRQHGYEVSQDVFKRFKDENGNFMESLSKDTKGLLSLYEASYFSFDGE  240
Ro_38431.3      IGVHAIALCFRLCRQHGYEVSQDVFKRFKDENGNFMESLSKDTKGLLSLYEASYFSFDGE  208
                ************************************************************

Ro_38431.2      QLMEEAKVFTAKHLKGKLANKDLVEQINHALEMPLQHRMLRLEARWYIEAYGKRKDANYL  290
Ro_38431.1      QLMEEAKVFTAKHLKGKLANKDLVEQINHALEMPLQHRMLRLEARWYIEAYGKRKDANYL  300
Ro_38431.3      QLMEEAKVFTAKHLKGKLANKDLVEQINHALEMPLQHRMLRLEARWYIEAYGKRKDANYL  268
                ************************************************************

Ro_38431.2      LLEMAMLEFNMVQSMLQGELKDMSRWWEDIDLGKRLSFTRDRLMECYFWNVGLIFEPKFS  350
Ro_38431.1      LLEMAMLEFNMVQSMLQGELKDMSRWWEDIDLGKRLSFTRDRLMECYFWNVGLIFEPKFS  360
Ro_38431.3      LLEMAMLEFNMVQSMLQGELKDMSRWWEDIDLGKRLSFTRDRLMECYFWNVGLIFEPKFS  328
                ************************************************************

Ro_38431.2      DCRKSLTKLTVFITTIDDVYDVYGSLDELELFTAAIHRWDIEAVETLPDYMKLCFLALYN  410
Ro_38431.1      DCRKSLTKLTVFITTIDDVYDVYGSLDELELFTAAIHRWDIEAVETLPDYMKLCFLALYN  420
Ro_38431.3      DCRKSLTKLTVFITTIDDVYDVYGSLDELELFTAAIHRWDIEAVETLPDYMKLCFLALYN  388
                ************************************************************

Ro_38431.2      TTNEMAYDILKRKGVNIIPHLKRAWADICKTFLMEAKWCSNRETPTFKAYLDNALISVSG  470
Ro_38431.1      TTNEMAYDILKRKGVNIIPHLKRAWADICKTFLMEAKWCSNRETPTFKAYLDNALISVSG  480
Ro_38431.3      TTNEMAYDILKRKGVNIIPHLKRAWADICKTFLMEAKWCSNRETPTFKAYLDNALISVSG  448
                ************************************************************

Ro_38431.2      VLILVHVYFLLTETITQDALECLEKKYHPLVECSSLIFRFSNDLATSKAELERGESANSI  530
Ro_38431.1      VLILVHVYFLLTETITQDALECLEKKYHPLVECSSLIFRFSNDLATSKAELERGESANSI  540
Ro_38431.3      VLILVHVYFLLTETITQDALECLEKKYHPLVECSSLIFRFSNDLATSKAELERGESANSI  508
                ************************************************************

Ro_38431.2      LCYMHETGVSEQEARKHISSFIEEMWKKMNKERVAADSPFEKPFIETAFNLARIAQCTYQ  590
Ro_38431.1      LCYMHETGVSEQEARKHISSFIEEMWKKMNKERVAADSPFEKPFIETAFNLARIAQCTYQ  600
Ro_38431.3      LCYMHETGVSEQEARKHISSFIEEMWKKMNKERVAADSPFEKPFIETAFNLARIAQCTYQ  568
                ************************************************************

Ro_38431.2      YGDGHGAPDNKAKNRVLSVIIEPITLVQRLQHRSNLLATVS 631
Ro_38431.1      YGDGHGAPDNKAKNRVLSVIIEPITLVQRLQHRSNLLATVS 641
Ro_38431.3      YGDGHGAPDNKAKNRVLSVIIEPITLVQRLQHRSNLLATVS 609
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B

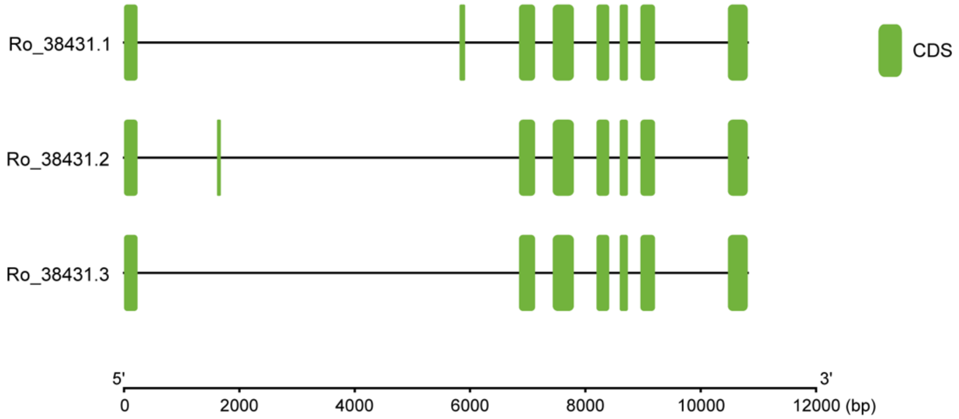
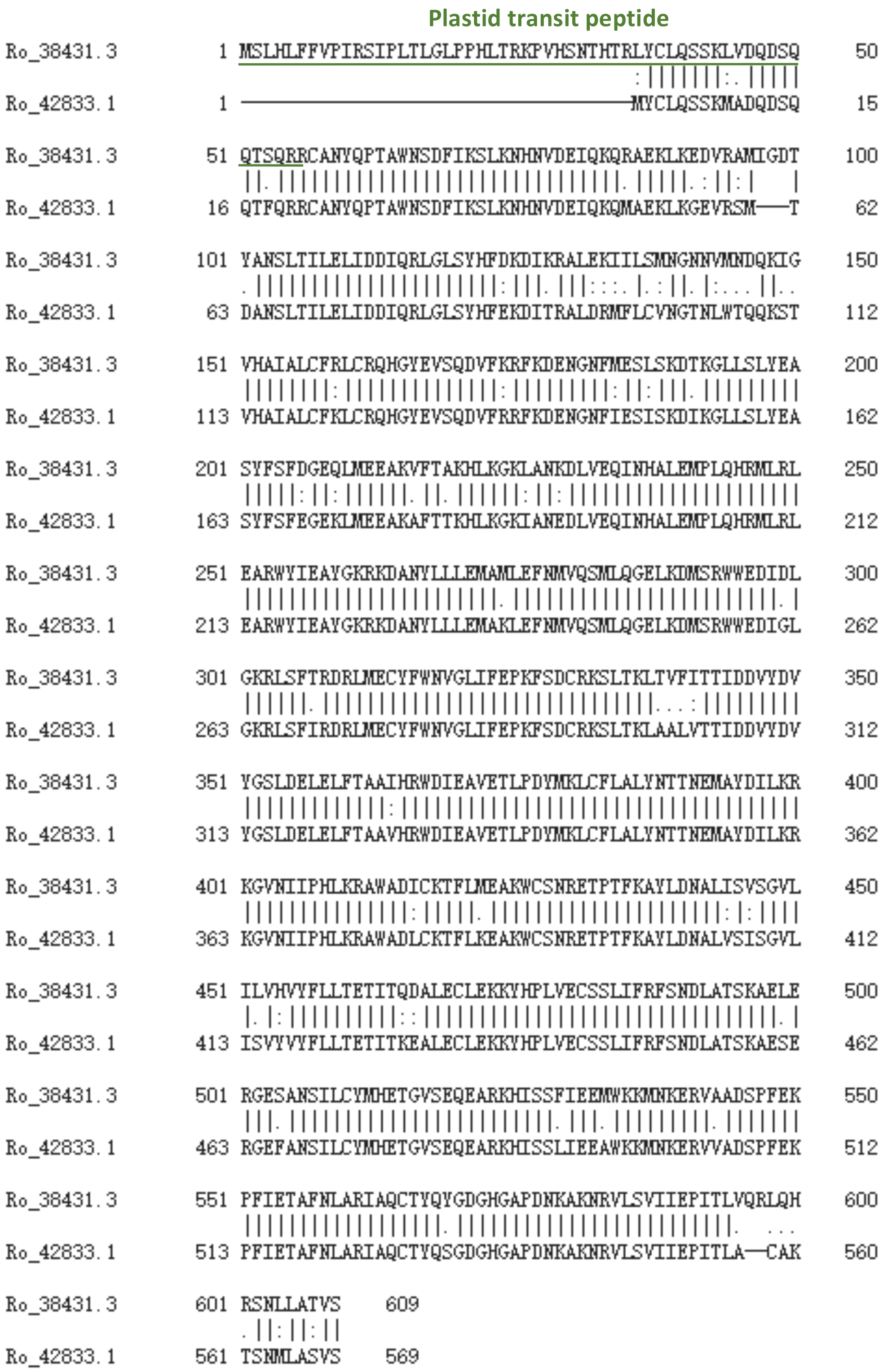


Figure S29 Structure features of the three alternative splicing transcripts of Ro_38431. A. Amino sequence alignment. B. gene structure.



```
Ro_38431.3           1 MSLHLFFVPIRSIPLTLGLPPHLTRKPVHSNTHTRLYCLQSSKLVDQDSQ     50
                                                          :|||||||:.|||||
Ro_42833.1           1 -----------------------------------MYCLQSSKMADQDSQ     15

Ro_38431.3          51 QTSQRRCANYQPTAWNSDFIKSLKNHNVDEIQKQRAEKLKEDVRAMIGDT    100
                       ||.|||||||||||||||||||||||||||||||.|||||.:||:|   |
Ro_42833.1          16 QTFQRRCANYQPTAWNSDFIKSLKNHNVDEIQKQMAEKLKGEVRSM---T     62

Ro_38431.3         101 YANSLTILELIDDIQRLGLSYHFDKDIKRALEKIILSMNGNNVMNDQKIG    150
                       .||||||||||||||||||||||:|||.|||:::.|.:||.|:...||..
Ro_42833.1          63 DANSLTILELIDDIQRLGLSYHFEKDITRALDRMFLCVNGTNLWTQQKST    112

Ro_38431.3         151 VHAIALCFRLCRQHGYEVSQDVFKRFKDENGNFMESLSKDTKGLLSLYEA    200
                       ||||||||:||||||||||||||:|||||||||:||:|||.|||||||||
Ro_42833.1         113 VHAIALCFKLCRQHGYEVSQDVFRRFKDENGNFIESISKDIKGLLSLYEA    162

Ro_38431.3         201 SYFSFDGEQLMEEAKVFTAKHLKGKLANKDLVEQINHALEMPLQHRMLRL    250
                       |||||:||:||||||.||.||||||:||:|||||||||||||||||||||
Ro_42833.1         163 SYFSFEGEKLMEEAKAFTTKHLKGKIANEDLVEQINHALEMPLQHRMLRL    212

Ro_38431.3         251 EARWYIEAYGKRKDANYLLLEMAMLEFNMVQSMLQGELKDMSRWWEDIDL    300
                       |||||||||||||||||||||||.||||||||||||||||||||||||.|
Ro_42833.1         213 EARWYIEAYGKRKDANYLLLEMAKLEFNMVQSMLQGELKDMSRWWEDIGL    262

Ro_38431.3         301 GKRLSFTRDRLMECYFWNVGLIFEPKFSDCRKSLTKLTVFITTIDDVYDV    350
                       ||||||.||||||||||||||||||||||||||||||...:|||||||||
Ro_42833.1         263 GKRLSFIRDRLMECYFWNVGLIFEPKFSDCRKSLTKLAALVTTIDDVYDV    312

Ro_38431.3         351 YGSLDELELFTAAIHRWDIEAVETLPDYMKLCFLALYNTTNEMAYDILKR    400
                       |||||||||||||:||||||||||||||||||||||||||||||||||||
Ro_42833.1         313 YGSLDELELFTAAVHRWDIEAVETLPDYMKLCFLALYNTTNEMAYDILKR    362

Ro_38431.3         401 KGVNIIPHLKRAWADICKTFLMEAKWCSNRETPTFKAYLDNALISVSGVL    450
                       |||||||||||||||:|||||.|||||||||||||||||||||:|:||||
Ro_42833.1         363 KGVNIIPHLKRAWADLCKTFLKEAKWCSNRETPTFKAYLDNALVSISGVL    412

Ro_38431.3         451 ILVHVYFLLTETITQDALECLEKKYHPLVECSSLIFRFSNDLATSKAELE    500
                       |.|:||||||||||::||||||||||||||||||||||||||||||||.|
Ro_42833.1         413 ISVYVYFLLTETITKEALECLEKKYHPLVECSSLIFRFSNDLATSKAESE    462

Ro_38431.3         501 RGESANSILCYMHETGVSEQEARKHISSFIEEMWKKMNKERVAADSPFEK    550
                       |||.||||||||||||||||||||||||.|||.|||||||||.|||||||
Ro_42833.1         463 RGEFANSILCYMHETGVSEQEARKHISSLIEEAWKKMNKERVVADSPFEK    512

Ro_38431.3         551 PFIETAFNLARIAQCTYQYGDGHGAPDNKAKNRVLSVIIEPITLVQRLQH    600
                       ||||||||||||||||||.|||||||||||||||||||||||||.  ...
Ro_42833.1         513 PFIETAFNLARIAQCTYQSGDGHGAPDNKAKNRVLSVIIEPITLA--CAK    560

Ro_38431.3         601 RSNLLATVS    609
                       .||:||:||
Ro_42833.1         561 TSNMLASVS    569
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Figure S30 Amino sequence alignment of Ro_38431.3 and Ro_43236.1. As an example, shows the location of plastid transit peptide.

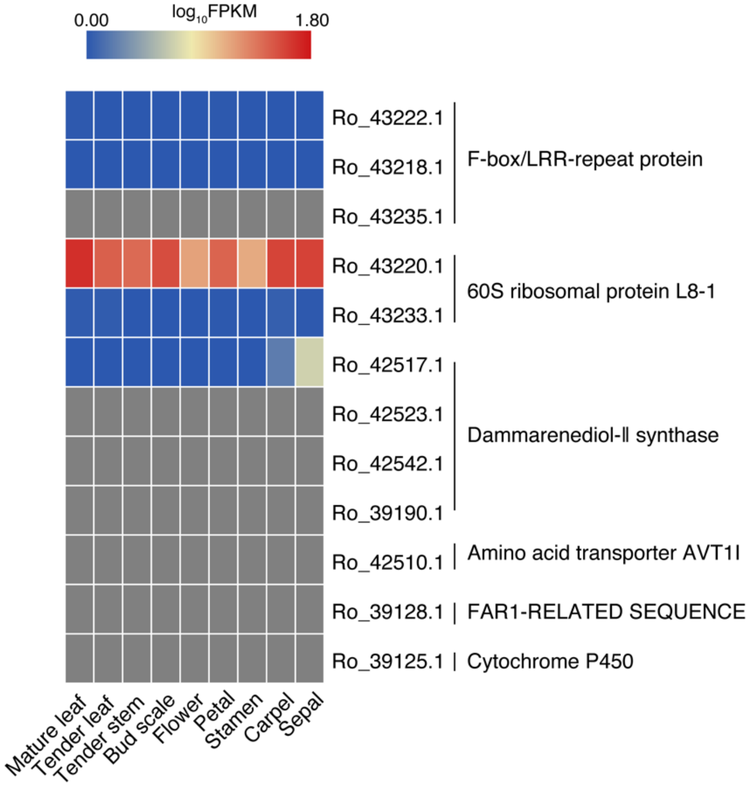

Figure S31 Expression profiles of adjacent genes of *TPS* clusters on chromosome 7. The gray blocks indicate no expression was detected.