

A)

```
1      10      20      30      40      50      60      70
AaTPS1 MSLSVGFGIGIPSS-PLASNAIRLVSSSYA-QANLPNHRVHMRTFRCTISEPFOITNDOKIDRRSAFYPSIWDYNYV
AcTPS1 MSLSVGFGGLVMPSS-LIASNAIRLVSSHASQAKLPVHORVRIILRCRTAEPSONDOKIDRRSAFYPSIWDYNYV
CIPIN  MAINLLSSIPAA CNFTRLSLPLSSKLVNG-----FWPPITRVOYHVAASTTPIKPVDOITIRRSADYPTIWSFDYI
CITEP  MAINLLSSIPAA CNFTRLSLPLSSKLVNG-----FWPPITVOYVMAASTSSIKPVDOITIRRSADYPTIWSFDYI
VvTER  MAISMLSSIPN--LITHTRIPITIKSSS-----CKASPRGKIKVKIGN--SNCBEITVRRRTANYEPTIWDYDYV

80      90      100     110     120     130     140     150
AaTPS1 OSLSKYAGDMYVKWAAMLKEDVVKRMMDV-----VDPIDGLGLITDDHORLGVYHFEDETKRITLESYINHINBGNW-
AcTPS1 OSLSNRYAGDMYVKWATMLKEDVVKRMMDKV-----VDPIDGLGLITDDHORLGVYHFEDETKRITLESYINHIBGNW-
CIPIN  OSLSKYKGGESYAROLEKLEKQVSAVMDQDNKVVDFDITLHOLELITDNIHRLGVSYHFEDETKRITLDRITL--NKNTN-
CITEP  OSLSKYKGGESYAROLEKLEKQVSAVMDQDNKVVDFDITLHOLELITDNIHRLGVSYHFEDETKRITLDRITL--NKNTN-
VvTER  OSLSRSDYVGETYTRRLDKLKRQVSAVMDQDNKVVDFDITLHOLELITDNIHRLGVSYHFEDETKRITLDRITL--NKNTN-

160     170     180     190     200     210     220     230
AaTPS1 TDDEHQAATSLKFRLLROHGYTIS-EDVFKNEKDEIGNPK-TCCLCEDVGLLILYEASYFSIEGER-LEEARDEFTKKN
AcTPS1 TDDEHQAATSLKFRLLROHGYTIS-EDVFKNEKDEIGNPK-TCCLCEDVGLLILYEASYFSIEGER-LEEARDEFTKKN
CIPIN  --KSIYATALKFRITROYGYNTVVKETISREMDKGSFKS--SHSDDCKGMLALYEAAYLVEEES--FRDAKSFMTAY
CITEP  --KSIYATALKFRITROYGYNTVVKETISREMDKGSFKS--SHSDDCKGMLALYEAAYLVEEES--FRDAKSFMTAY
VvTER  QKDDIYVALEERLLROHGYDVP-EDVFSREKDDIGSEK-ACCLCEDMKGMLCLYEASYMCOGES-TMEQARDEFAHRH

240     250     260     270     280     290     300     310
AaTPS1 LKK--TLKRKDMIDDDI--IAITLVSHAFELPHWRVRFEARWFIDVYERRRPNLSPITLITAKLDENMVOAHOEDLKHV
AcTPS1 LNK--TLKRKDMIDDDI--IAITLVSHAFELPHWRVRFEARWFIDVYERRRPNLSPITLITAKLDENMVOAHOEDLKHV
CIPIN  LKQVWVTEHDNKKHDDHITCITLVNHAFELPHWRMRFEARWFIDVYENGPDMNPITLITAKVDENIVQAVHOENLKYA
CITEP  LKQVWVAKHDIDKNDNBYITCITLVKHAFLPHWRMRFEARWFIDVYBSPDMNPITLITAKVDYNIQVAVHOEDLKYV
VvTER  LG---KGLEQNTDQN--IAITLVKHAFLPHWRMRFEARWFIDVYBKRQDMNPITLITAKLDENMVOAHOEDLRHM

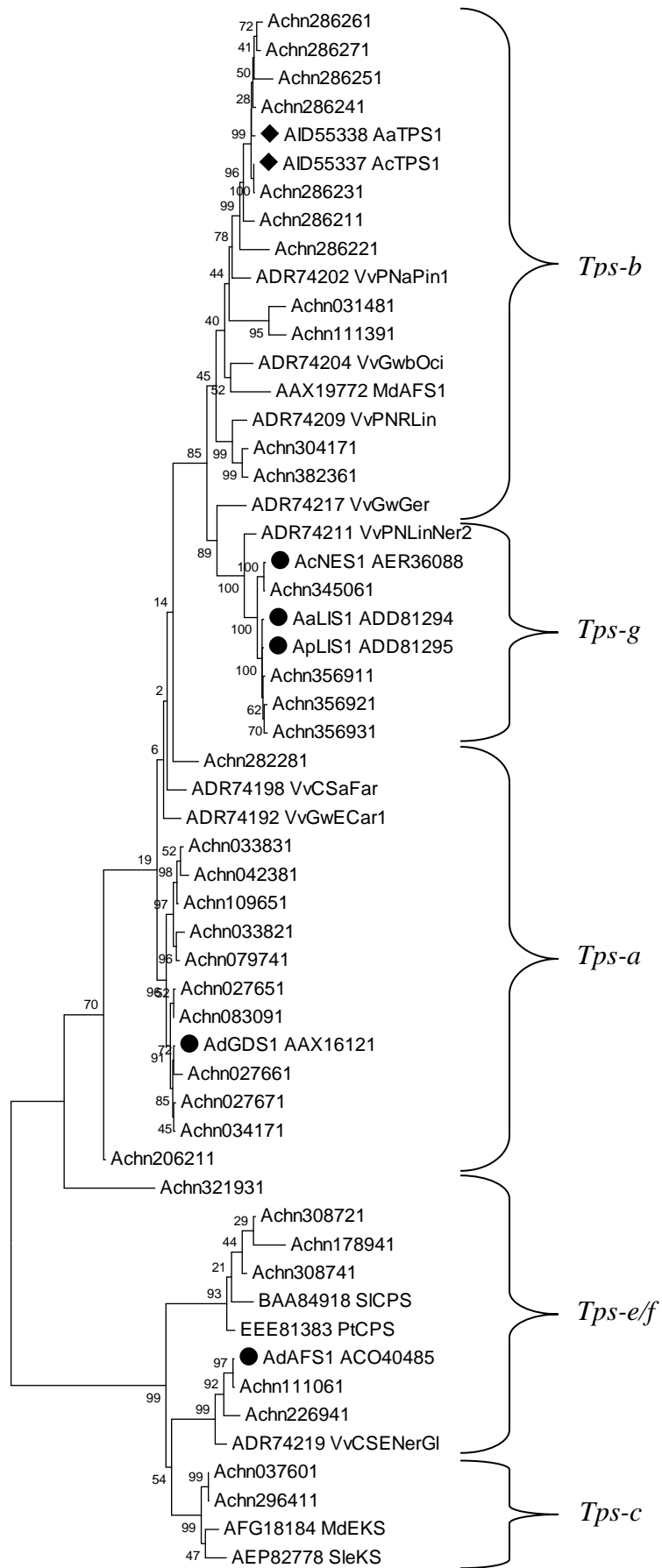
320     330     340     350     360     370     380     390
AaTPS1 SRWVNNRILGOKLSFARDRLMENEFTWVGMDKPRSYERKNMILVNSITLITIDDDVYDVYGTLDLELEFETNAVERWDL
AcTPS1 SRWVNNRILGOKLSFARDRLMENEFTWVGMDKPRSYERKNMILVNSITLITIDDDVYDVYGTLDLELEFETNAVERWDL
CIPIN  SRWVKKKILGKLNFRDRIVENEFWVYGEKFEPOGCFYRRMSIMVNAITLAVDDVYDVYGTLDLELEFETDAVERWDA
CITEP  SRWVKKKILGKLNFRDRIVENEFWVYVGDIFEEPOGCFYRRMSAMVNCITLITIDDDVYDVYGTLDLELEFETDAVERWDA
VvTER  SSWVSSRILGKLNFRARDRLMENEFTWVVGVIIFEPQYCFYRRMSIKVNTLITIDDDVYDVYGTLDLELEFETDVVDRWDL

400     410     420     430     440     450     460
AaTPS1 SEMBHIIPAYMKICFLATFNSINEMA YDTIKEOGVHITP-YLQKMWADICKSYLVEAKWYVSRHTPSFPEYMNNAWISI
AcTPS1 SEMBHIIPAYMKICFLATFNSINEMA YDTIKEOGVHITP-YLQKMWADICKSYLVEAKWYVSRHTPSFPEYMNNAWISI
CIPIN  TAVBQIIPAYMKICFLATFNSINEMA YDIALRDOGVITPISYLTAKAWADICKAYLVEAKWYNSGYIPPLPEYMNNAWISI
CITEP  TITBQIIPAYMKICFLATFNSINEMA YDIALRDOGVITPISYLTAKAWADICKSYLVEAKWYNSGYIPPLPEYMNNAWISV
VvTER  NAMDPIIPAYMKICFLATFNSINEMA YDALKHEGLHITVSYLRKAWSDICKSYLLEAKWYVSRHTPSFPEYISNSWISI

470     480     490     500     510     520     530     540
AaTPS1 SASVMLAHAYVLCNTNITSYEGLKQVQKYPNLRCSAIIIRLADDLATSIDEMERGDIPKSTOCYMHETGISEEARKH
AcTPS1 AAPVALAHAYVLCNTNITAKEGLKQVQKYPNLRCSAIIIRLADDLATSIDEMERGDIPKSTOCYMHETGASEKDKARKH
CIPIN  GATVMLVHANTFTANETIKKEGLEFVKQVYENLRSSMIIIRLADDLATSIDELKRGDVPKSTOCYMHETGISEGEAREH
CITEP  TAPVMLLHAYVFTANETIKKEALEFLQSDIIRISSMIIIRLADDLATSIDELKRGDVPKSTOCYMHETGISEGEAREH
VvTER  SGPVMLVHAYVIVANETIKKEALQSLEIRVHNIIRSSMIIIRLSDDLATSIDELKRGDVPKSTOCYMYETGASEEDARKH

550     560     570     580     590     600     610     620
AaTPS1 MKYVITCEALKKMNEARVEDGPFPSRAITGVVENIVRMOCMYOYGDGHC-AQGPETTRDRVKSLLINPITFEFEKA
AcTPS1 MKYVITCEALKKMNEARVEDGSLFSRTITGVAENIARMSOCMYOYGDGHC-AQGPETTRDRVKSLLINPITLFLF
CIPIN  INDIITAOITWMMKMRDRFNGPHFVSDVEVGIAMNLRMSOCMYOYGDGHC-CGAQETTRARVLSLFFDPTA
CITEP  IIRDIITAOITWMMKMSARFGNPPYLPDVEITGIAMNLRMSOCMYLGDGHC--GVQENTTRDRVLSLFFDPTI
VvTER  TSNVITCEATWKKLNEDGAVESPEPEITIGIAMNLRMAOCMYOHGDGHC-IEYGFETTRDRVLSLFFVPTISLSSE
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B)



1

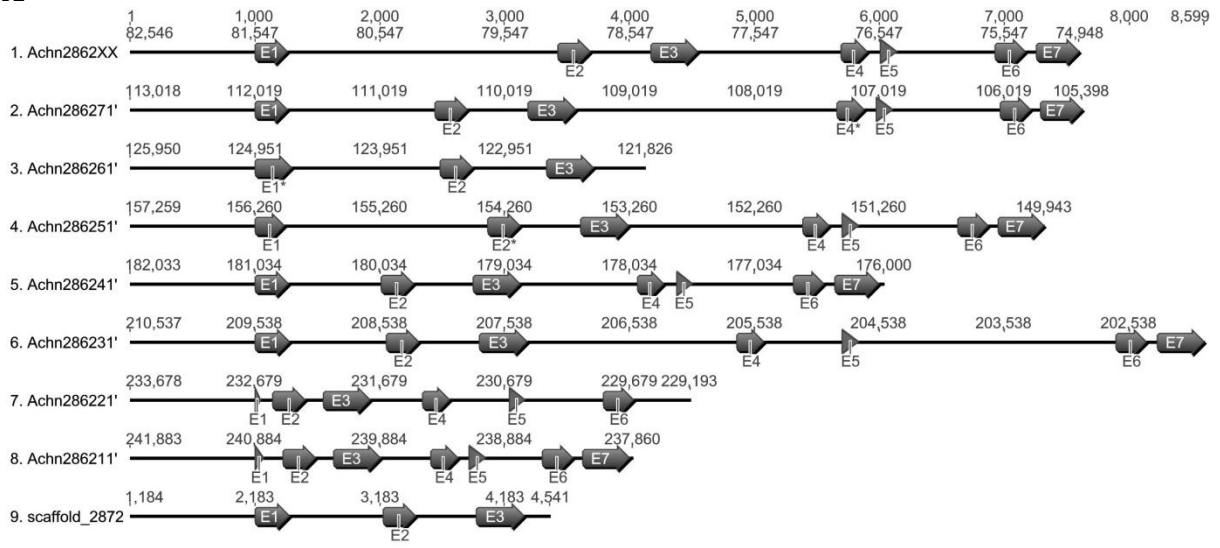
### Figure S1.

(A) Alignment of the deduced amino acid sequences of *AaTPS1* and *AcTPS1* and other *Tps-b* subfamily terpene synthases. Abbreviations with GenBank accessions in brackets: *AaTPS1* (*Actinidia arguta* terpene synthase 1 - AID55338), *AcTPS1* (*A. chinensis* terpene synthase 1 - AID55337), CIPIN (*Citrus limon* (-)- $\beta$ -pinene synthase - AAM53945), CITEP (*Citrus limon*  $\gamma$ -terpinene synthase - AAM53943) and VvTER (*Vitis vinifera* (-)- $\alpha$ -terpineol synthase - AAS79351). The highly conserved metal cofactor binding region (DDXXD) as well as the conserved RXR and RRX<sub>8</sub>W motifs are underlined. The cleavage sites of the predicted chloroplast targeting peptides (ChloroP predictions) are marked with arrowheads. Sequence alignments were constructed using ClustalX (Thompson, 1997) and visualized using MEGA5 (Tamura et al., 2011).

(B) Unrooted Maximum Likelihood tree of *AcTPS1* and *AaTPS1* with other terpene synthases from kiwifruit, grape, apple, tomato and poplar. *AcTPS1* and *AaTPS1* from this work are indicated by black diamonds. Previously published functionally characterized kiwifruit TPS sequences are indicated by black circles. Thirty-five TPS gene models (Achn numbers) were identified in the kiwifruit 'Hongyang' genome (Huang et al., 2013) using BLASTP searches (cut-off < 1e<sup>-2</sup>) using full length TPS protein sequences from grape, tomato, apple, poplar as query sequences. The evolutionary history was inferred by using the Maximum Likelihood method based on the Dayhoff matrix based model (Schwarz and Dayhoff, 1979). The percentage of trees in which the associated taxa clustered together is shown next to the branches. Initial tree(s) for the heuristic search were obtained automatically by applying Neighbor-Join and BioNJ algorithms to a matrix of pairwise distances estimated using a JTT model, and then selecting the topology with superior log likelihood value. The tree is drawn to scale, with branch lengths measured in the number of substitutions per site. The analysis involved 55 amino acid sequences. There were a total of 2260 positions in the final dataset. Evolutionary analyses were conducted in MEGA6 (Tamura et al., 2013). Multiple amino acid sequence alignments of the TPS genes were first performed with ClustalW using the using Geneious® v6.1.6 (www.geneious.com), the alignment was then exported in Phylip format and imported in MEGA6 for the analysis. Bootstrap values are shown as a percentage based on 1000 replicates. The scale bar represents expected changes per site. TPS families are bases on (Bohlmann et al., 1998; Dudareva et al., 2003).

Abbreviations with GenBank accessions in brackets are as follows: *AaTPS1* = *Actinidia arguta* terpene synthase 1 (AID55338); *AcTPS1* = *A. chinensis* terpene synthase 1 (AID55337); *AdAFS1* = *A. deliciosa* (E,E)- $\alpha$ -farnesene synthase (ACO40485); *AdGDS1* = *A. deliciosa* (+)-germacreneD synthase (AAX16121); *AaLIS1* = *A. arguta* linalool synthase (ADD81294); *ApLIS1* = *A. polygama* linalool synthase (ADD81295); *AcNES1* = *A. chinensis* nerolidol synthase (AER36088); *VvGwECar1* = *Vitis vinifera* (E)- $\beta$ -caryophyllene synthase (ADR74192), *VvCSaFar* = (E,E)- $\alpha$ -farnesene synthase (ADR74198), *VvPNRLin* = (3R)-linalool synthase (ADR74209), *VvPNaPin1* = pinene synthase (ADR74202), *VvGwbOci* = (E)- $\beta$ -ocimene synthase (ADR74204), *VvGwGer* = geraniol synthase (ADR74217), *VvPNLinNer2* = (3S)-linalool/(E)-nerolidol synthase (ADR74211), *VvCSEnerG1* = P(E)-nerolidol/(E,E)-geranyl linalool synthase (ADR74219); *MdAFS1* = *Malus domestica* (E,E)- $\alpha$ -farnesene synthase (AAX19772), *MdEKS* = ent-kaurene synthase (AFG18184); *SleKS* = *Solanum lycopersicum* ent-kaurene synthase (AEP82778), *SICPS* = copalyl diphosphate synthase (BAA84918); *PtCPS* = *Populus trichocarpa* copalyl diphosphate synthase (EEE81383).

A





C

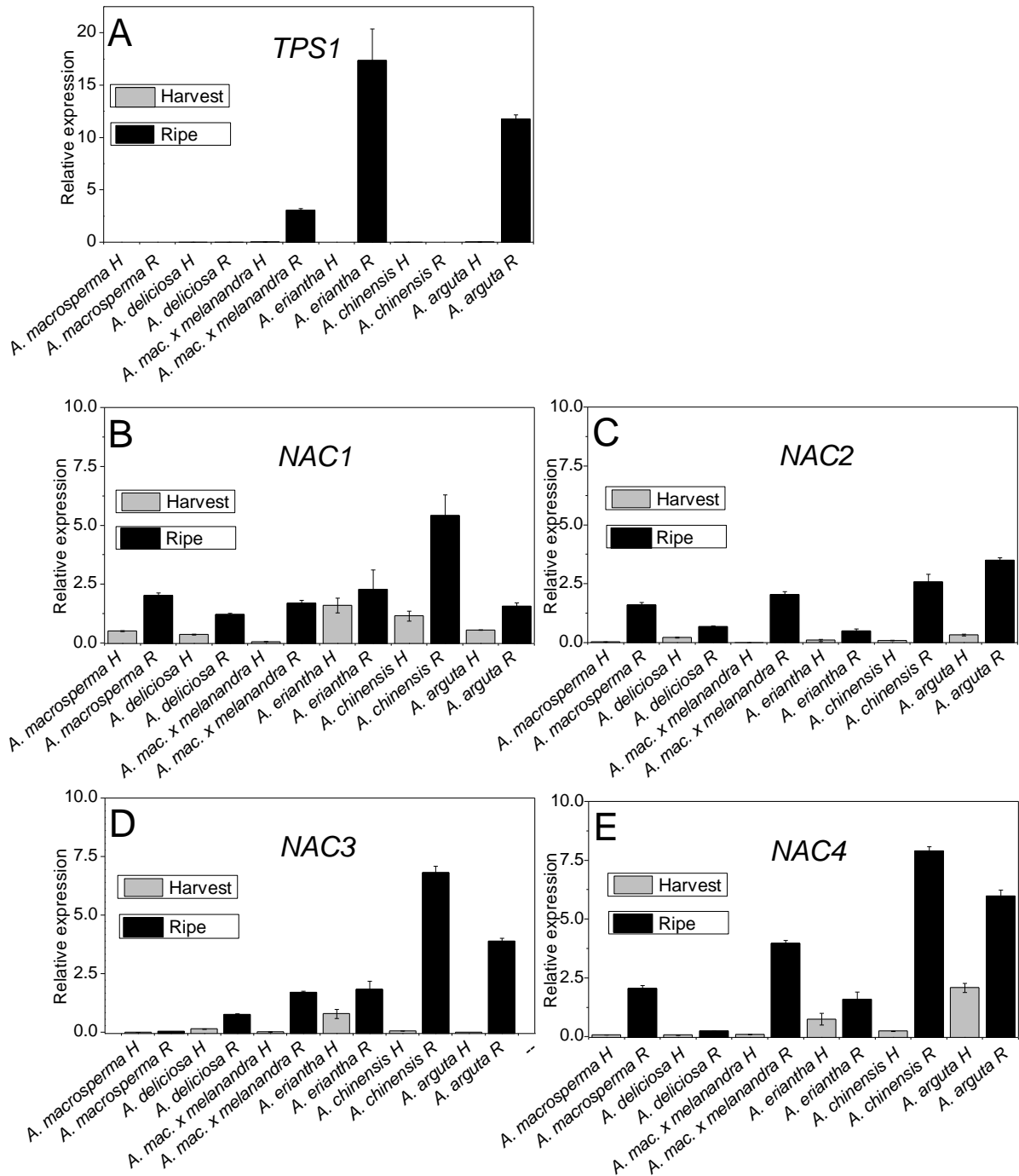
	Achn286261'	scaffold_2872	Achn286231'	AcTPS1	Achn2862XX	Achn286271'	Achn286241'	Achn286251'	AaTPS1	Achn286211'	Achn286221'
Achn286261'		80.1%	80.1%	80.4%	80.9%	86.6%	82.0%	95.4%	80.3%	71.7%	68.5%
scaffold_2872	80.1%		93.0%	93.4%	88.0%	87.7%	86.4%	93.7%	87.4%	74.2%	69.5%
Achn286231'	80.1%	93.0%		98.8%	87.9%	88.6%	87.1%	87.0%	87.6%	80.3%	72.1%
AcTPS1	80.4%	93.4%	98.8%		88.4%	89.2%	87.7%	87.8%	88.2%	80.6%	73.1%
Achn2862XX	80.9%	88.0%	87.9%	88.4%		89.6%	86.0%	86.7%	86.5%	79.5%	72.6%
Achn286271'	86.6%	87.7%	88.6%	89.2%	89.6%		89.4%	91.3%	89.3%	81.2%	74.3%
Achn286241'	82.0%	86.4%	87.1%	87.7%	86.0%	89.4%		90.4%	88.4%	78.8%	73.6%
Achn286251'	95.4%	93.7%	87.0%	87.8%	86.7%	91.3%	90.4%		87.9%	77.1%	72.2%
AaTPS1	80.3%	87.4%	87.6%	88.2%	86.5%	89.3%	88.4%	87.9%		81.1%	73.7%
Achn286211'	71.7%	74.2%	80.3%	80.6%	79.5%	81.2%	78.8%	77.1%	81.1%		79.8%
Achn286221'	68.5%	69.5%	72.1%	73.1%	72.6%	74.3%	73.6%	72.2%	73.7%	79.8%	

**Figure S2.**

(A) Mapping of *AcTPSI* exons 1-7 onto scaffolds in the kiwifruit genome. To improve the TPS gene models identified in the kiwifruit genome, exons 1-7 of *AcTPSI* were used as BLASTn query sequences against the kiwifruit genome scaffold database (<http://bioinfo.bti.cornell.edu/cgi-bin/kiwi/home.cgi>). A schematic showing the structure of eight tandemly repeated, full-length and partial TPS1-like gene models (Achn286211'-XX) identified on scaffold\_575 (length 316,842 bp) is shown. Scaffold\_2872 (length 5307 bp) also contained a portion homologous to *AcTPSI* exons 1-3.

(B) Amino acid alignment of manually curated kiwifruit TPS1-like gene models. Exons 1-7 for each TPS1-like gene model were extracted from the kiwifruit scaffold database, joined together, translated into protein and aligned using Geneious® v6.1.6/ClustalW/default settings ([www.geneious.com](http://www.geneious.com)). Sequences were manually curated to maximise alignment and the designated with a prime (') symbol. Only three genes (Achn286231', Achn286241' and Achn2862XX) showed an intact structure of seven complete exons and no in-frame stop codons (asterisks). The conserved RRX8W and DDXXD domains are boxed.

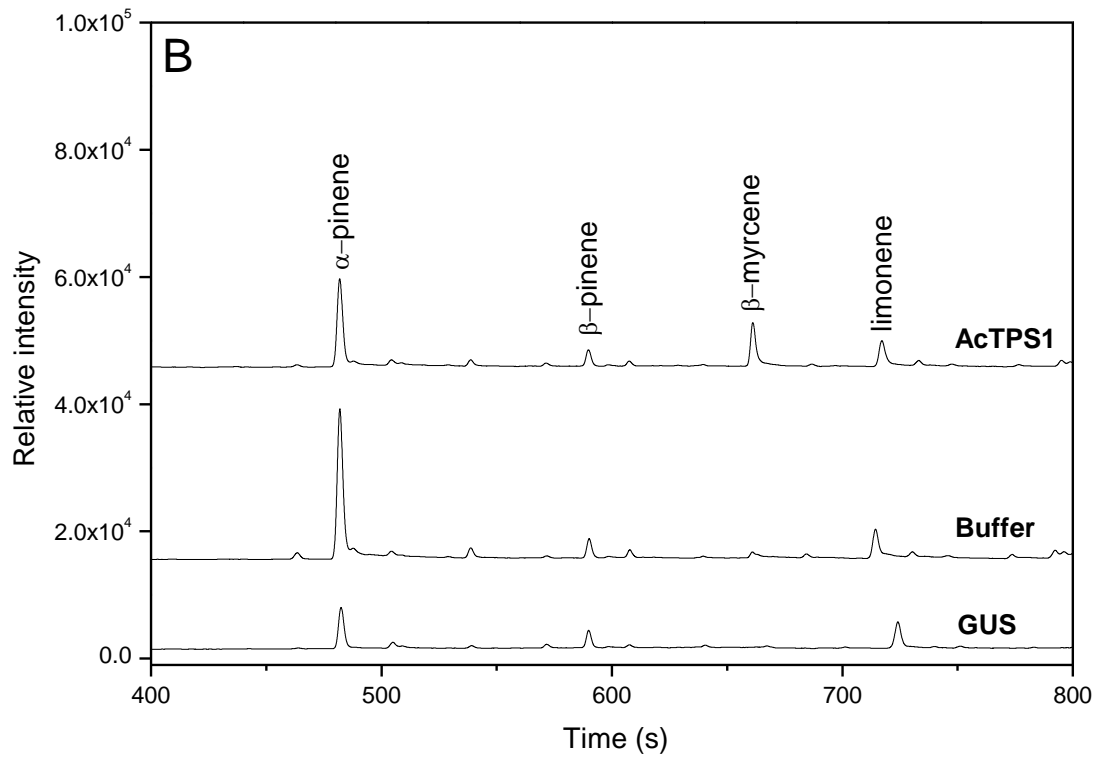
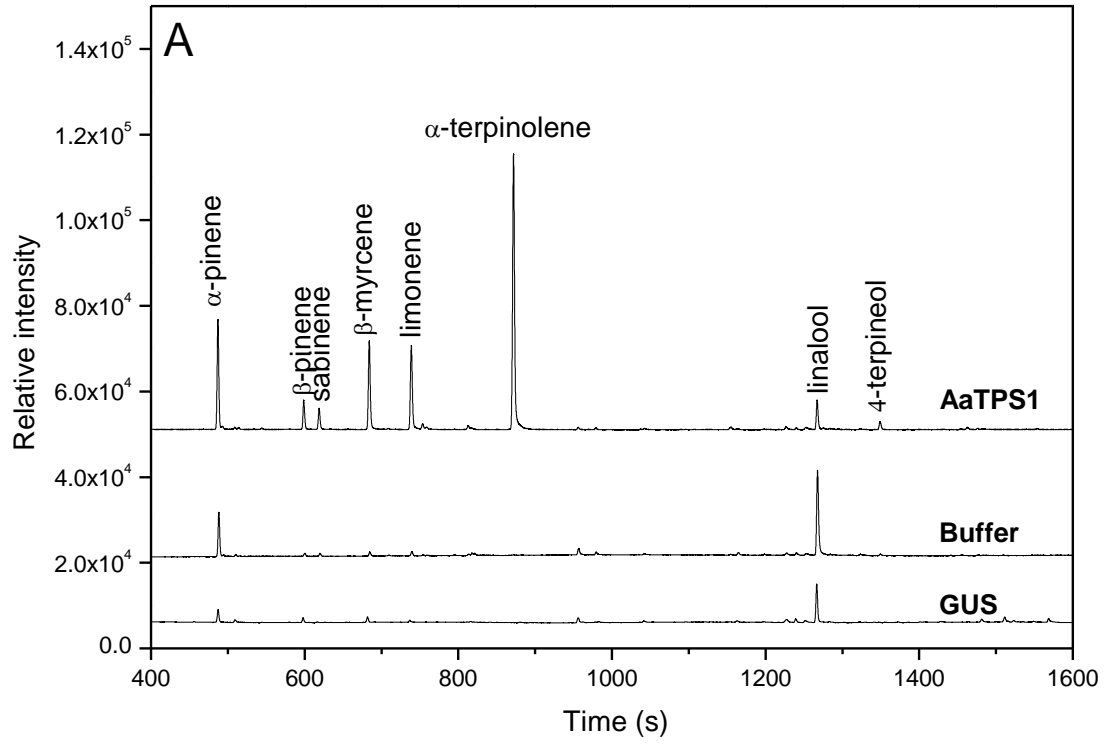
C) Sequence identity table of aligned kiwifruit TPS1-like amino acid sequences.



**Figure S3.**

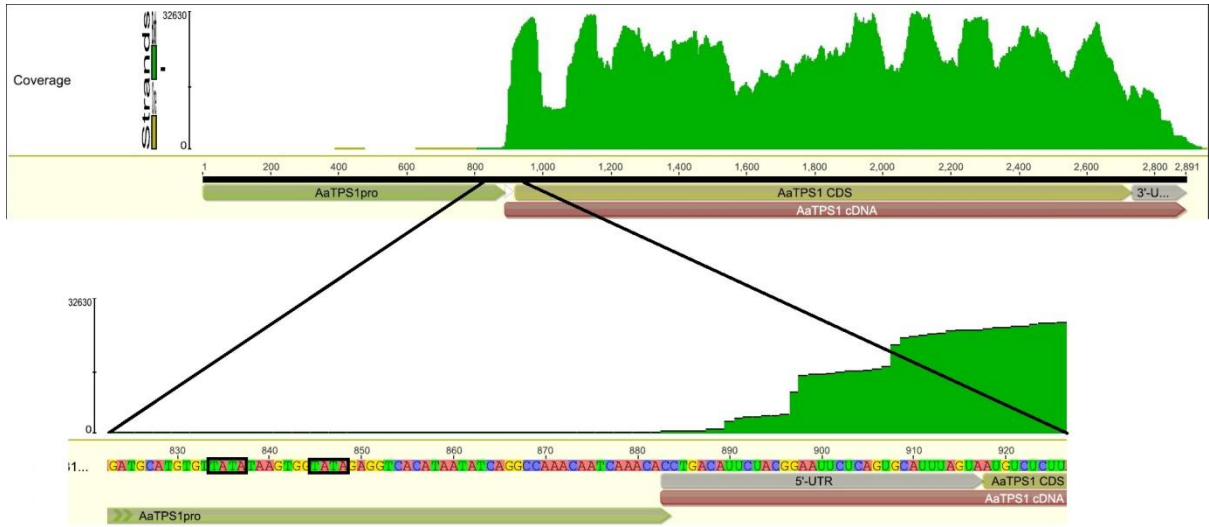
Real-time qPCR analysis of *TPS1* (A) and *NAC1-4* (B-E) gene expression levels in harvest (H) and ripe (R) fruit for six *Actinidia* species. Analyses were performed on pooled fruit samples using gene specific primers given in Table S2. The *TPS1* primers were designed to amplify both *AaTPS1* and *AcTPS1*. *Achn286241* and *Achn2862XX* expression was also tested using gene specific primers but both were not expressed in harvest or ripe fruit. Error bars are based on four technical replicates and data were normalized against the housekeeping gene *EF1 $\alpha$* . The *A. mac. x melanandra* sample represent fruit from an interspecific cross between *A. macrosperma* and *A. melanandra*.





**Figure S4.** GC-MS analysis of the monoterpenes produced by transient expression of *AaTPS1* and *AcTPS1* in *planta*. The complete open reading frames of *AaTPS1* and *AcTPS1* were cloned into the binary vector pHEX2 and transformed into *Agrobacterium tumefaciens*. Bacterial cell suspensions were injected into the abaxial side of three young *Nicotiana benthamiana* leaves and volatiles were assayed 2 weeks after injection by dynamic headspace sampling of detached leaves. Leaves were infiltrated with buffer-only and with a vector expressing the *GUS* reporter gene (pHEX2-*GUS*) as controls. (A) *AaTPS1* - upper trace; buffer-only (mock infiltrated) - middle trace; *GUS* - lower trace. (B) *AcTPS1* - upper trace; buffer-only - middle trace; *GUS* - lower trace. Analyses were made in triplicate with only a single representative trace shown for each treatment.

A





### Figure S5.

(A) Identifying the transcriptional start site in the 5'-UTR of *AaTPS1*. Illumina sequencing data derived from *Actinidia arguta* fruit mRNA (ripening stage C3, Fig 2) was read mapped against the *AaTPS1* gene (*AaTPS1* promoter fused to the *AaTPS1* cDNA). Boxed sequences = putative TATA boxes. The 5'-UTR (in grey) is estimated to be 35 bp from the ATG (start of translation). Read mapping was performed using Geneious® v6.1.6/Align/Assemble/Map to reference. Parameters: Custom, Word length 30, Index word length 15, ignore words repeated more than 12 times, Max mismatch per read 10%, Maximum ambiguity 4, No gaps allowed. Library size: 37,154,393 reads (41-88 bp post trimming).

(B) Sequence alignment of *AaTPS1*, *AcTPS1* and *Achn286241* promoter regions. The kiwifruit genome scaffold database (<http://bioinfo.bti.cornell.edu/cgi-bin/kiwi/home.cgi>) was searched using BLASTn to identify regions of homology to the *AaTPS1* promoter. The *AaTPS1* promoter sequence (~0.9 kb) was then aligned with the *AcTPS1* (~0.9 kb) and *Achn286241* (~2.2 kb) upstream regions using Geneious® v6.1.6/Muscle align/default settings ([www.geneious.com](http://www.geneious.com)). The *AaTPS1* promoter shares significant sequence identity to the *AcTPS1* (68%) and *Achn286241* promoter (71%, excluding insertion) but not to other TPS1-like genes on scaffold\_575. *Achn286241* shows an insertion of ~1.2 kb around the predicted TATA region (boxed), while *AcTPS1* shows significant divergence in the predicted NAC binding site (underlined at bp 750). The 5'-UTR in *AaTPS1* is underlined upstream of the ATG start codon (as determined above). The *AcTPS1* promoter sequence used in this work is 99% identical to the 3 kb *Achn286231* upstream region.

AaTPS1pro-668 1 GCTCCTCCTGAGCTTACGTTGGTTCCACGACAGGCAATGGAGCCAACGGAAAAATAAAATC 60  
GCT CTC TG GC TAC TTGGTTCCACGACAGGCA TGGAGCCAACGGAAAA AAAAT  
AcTPS1pro-648 1 GCTTCTCATGGGCATACATTGGTTCCACGACAGGCAGTGGAGCCAACGGAAAAAAAATG 60  
  
AaTPS1pro-668 61 CTAAGGAAAAATAAAATCAAACACTAGTACTAGCTAGGGTAAATCATGCCTTAAAAACTAA 120  
CT AGGAAAAATAAAA CAAACACTA TACTAG TAGG TAAATC GCC T AAA AA  
AcTPS1pro-648 61 CTTAGGAAAAATAAAACCAAACACTAATACTAGTTAGGATAAATC--GCCCTGAAATTCOA 118  
  
AaTPS1pro-668 121 GGCTATGTTTGACTATGTGAATTTAGATTTGAAGATTTCAAAATTC-----AGT 169  
CT TTTGA TA GTGAA TTAGATTTGAA ATTT AAAT CC GT  
AcTPS1pro-648 119 CTCTCCATTTGATTACGTGAAATTTAGATTTGAAAATTTTAAATCCCAACAATTTTAGGT 178  
  
AaTPS1pro-668 170 CTTTATAGATTTCAAAATTCCTCTATTTGAATACATATTTTTTTTTAAAAAATAA---TTAA 226  
CTTTAT GATTT AATCCTC ATTT ATACATATTTTTTTTTAAAAAATAA A TTAA  
AcTPS1pro-648 179 CTTTATGGATTTTCAATCCTCCATTTAGATACATATTTTTTTTTAAAAAATAAATCTTAA 238  
  
AaTPS1pro-668 227 TTATATGAATTTTAAATGAACACATAGTTTAGAAAGAGAGACGAGAGGAAAGAAATGTG 286  
TTATATGAA TT AAT AACAC TA TTTA AA GAG GAGAC AGAG TG  
AcTPS1pro-648 239 TTATATGAA-TTCAATAAACACCTATTTTAAATGAGTGAGACAAGAG-----ATG 288  
  
AaTPS1pro-668 287 AGAGAGACGAGGAATAAGAGAAAAGAAGGTGAAGTGAATTAATTGAAATCTTTTAA 346  
A AGAGA A A GA GGTGAAGTGAATG AT ATTTGAA T TTTTAA  
AcTPS1pro-648 289 ACAGAGA-----AGATGAGGGTGAAGTGAATGAGATAAATTGAAATTTTTTTAA 335  
  
AaTPS1pro-668 347 TTTTACTTTTGAATTC AATAGGTAATTTTTATTTATTTTAAATTTTTTGTATCTATTCGC 406  
TTTT CT TT AATCAATAGG ATTT TATT ATTT AAAT TTTTGT TATTCGC  
AcTPS1pro-648 336 TTTTGTCTTGTAAATTC AATAGGCCATTTCTATTCATTTCAAAATCTTTTGTATTTATTCGC 395  
  
AaTPS1pro-668 407 CATCCCAACATACTCAACAGATTTGAGAGAAGAGTTTACGTAATTC A A A C A G A T T C C A A A G 466  
ATCC AACATACTC A GATTTGAGA AAGA T AC TAT CAAAC GA C AAAG  
AcTPS1pro-648 396 TATCCAAACATACTCGATGGATTTGAGACAAGAATCCACATAATCC A A A C G G A G C C T A A A G 455  
  
AaTPS1pro-668 467 TCACAAACCAAGGACCAACCTAGACCACATAAAAAAGACCAAGGACCACACACAAATTA 526  
TCACAAACCAAGGACCAA CTAGACCACATAAAAAAGACCAAGGACCA ACACAAATTA  
AcTPS1pro-648 456 TCACAAACCAAGGACCAATCTAGACCACATAAAAAAGACCAAGGACCATACACAAATTA 515  
  
AaTPS1pro-668 527 AGAGAAAACCCACACGCACACGCCCAAAAAAAGGGTGAAAATCTTCAATGTCATGTGTTA 586  
AGA AAAACCCAC CGCACA CAAAAAAA GG AAAATCTTGAGATGCATGTGTTA  
AcTPS1pro-648 516 AGAAAAAACCCACGCACACAAA--CAAAAAAAGGAAAAATCTTCAATGTCATGTGTTA 573  
  
AaTPS1pro-668 587 TATAAGTGGTATAGAGGTCACATAATATCAGGCCAAACAATCAAACACCTGACATTCTAC 646  
TATAA AGAGG CACATA TA CAGGCCAAACAA CAA CA CTGACATTCTA  
AcTPS1pro-648 574 TATAA-----AGAGGCCACATACTAGCAGGCCAAACAACCAAGCAGCTGACATTCTAA 626  
  
AaTPS1pro-668 647 GGAATTCTCAGTGCATTTAGCCATG 671  
GGAATTCTCAGTGCAT TAGCCATG  
AcTPS1pro-648 627 GGAATTCTCAGTGCATCTAGCCATG 651

**Figure S6.** Binding sites in the *AaTPS1* and *AcTPS1* promoter regions. The translational start codon is indicated in bold. Highlighted promoter elements are: EIN3 binding site **AYGWAYCT** (Kosugi and Ohashi, 2000); EIN3 binding site **ATGCATCT** (Boutrot et al., 2010); NAC binding site **YACGTAABY** (Fig 6); Ethylene Response Elements (EREs) **AWTTCAAA** (Montgomery et al., 1993; Itzhaki et al., 1994; Tapia et al., 2005). Underlined: 5'-untranslated region (5'-UTR) based on Fig. S5A (*AaTPS1*) or available EST information (*AcTPS1*).

A)

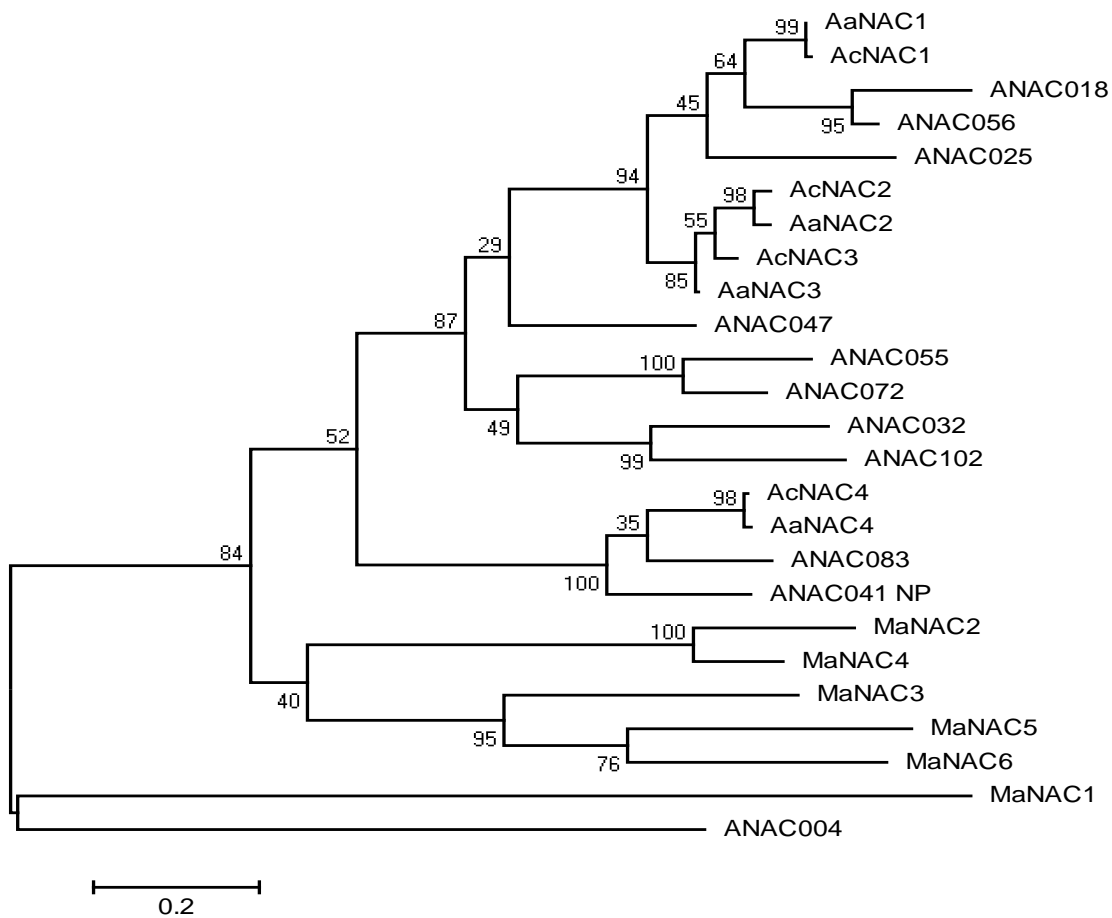
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1          10          20          30          40          50          60          70          80
Consensus  MFS TDSVSGSQQP I LPPGRRRHHFDDBFAVH YLKKKAASAPLPPVSTHIAVMDIYKFDPPWLPKAKATFGEOBMYEFSPRDRKYPN
AaNAC1     MFS TDSSTGSPQPI LPPGRRRHHFDDBFAVH YLKKKAASAPLPPVSTHIAVMDIYKFDPPWLPKAKATFGEOBMYEFSPRDRKYPN
AcNAC1     MFS TDSSTGSPQPI LPPGRRRHHFDDBFAVH YLKKKAASAPLPPVSTHIAVMDIYKFDPPWLPKAKATFGEOBMYEFSPRDRKYPN
AaNAC3     MFS TDSVSGSQQP I LPPGRRRHHFDDBFAVH YLKKKAASAPLPPVSTHIAVMDIYKFDPPWLPKAKAMFGEOBMYEFSPRDRKYPN
AaNAC2     MFS TDSVSGSQQP I LPPGRRRHHFDDBFAVH YLKKKAASAPLPPVSTHIAVMDIYKFDPPWLPKAKAMFGEOBMYEFSPRDRKYPN
AaNAC2     MFS TDSVGLQQP I LPPGRRRHHFDDBFAVH YLKKKAASAPLPPVATIAVMDIYKFDPPWLPKAKATFGEOBMYEFSPRNRKYPN
AaNAC4     MFS TDSVGLQQP I LPPGRRRHHFDDBFAVH YLKKKAASAPLPPVATIAVMDIYKFDPPWLPKAKATFGEOBMYEFSPRNRKYPN
AcNAC4     MKL N-VVKNVIR LPPGRRRHHFDDBFAVH YLKKKAASAPLPPVATIAVMDIYKFDPPWLPKAKATFGEOBMYEFSPRNRKYPN
AcNAC4     MKL N-VVKNVIR LPPGRRRHHFDDBFAVH YLKKKAASAPLPPVATIAVMDIYKFDPPWLPKAKATFGEOBMYEFSPRNRKYPN

90          100          110          120          130          140          150          160
Consensus  GARP NRAATSGYKATC DKKP VLTSSGSOKVGVKKAIVFYRCKPKGVKUNWIMHEYRIIDNEAKSKPP-GYDAGXNKGSLRI
AaNAC1     GARP NRAATSGYKATC DKKP VLTSSGSOKVGVKKAIVFYRCKPKGVKUNWIMHEYRIIDNEAKSKPP-GCDAANKKASLRI
AcNAC1     GARP NRAATSGYKATC DKKP VLTSSGSOKVGVKKAIVFYRCKPKGVKUNWIMHEYRIIDNEAKSKPP-GCDAANKKASLRI
AaNAC3     GARP NRAATSGYKATC DKKP VLTSSGSOKVGVKKAIVFYRCKPKGVKUNWIMHEYRIIDNEAKSKPP-EYDACHNKSLRI
AaNAC2     GARP NRAATSGYKATC DKKP VLTSSGSOKVGVKKAIVFYRCKPKGVKUNWIMHEYRIIDNEAKSKPP-GYDACHNKSLRI
AaNAC2     GARP NRAATSGYKATC DKKP VLTSSGSOKVGVKKAIVFYRCKPKGVKUNWIMHEYRIIDNEAKSKPP-GYDACHNKSLRI
AcNAC2     GARP NRAATSGYKATC DKKP VLTSSGSOKVGVKKAIVFYRCKPKGVKUNWIMHEYRIIDNEAKSKPPGVYDACHNKSLRI
AaNAC4     GNS NRAATSGYKATC DKKP VLTSSGSOKVGVKKAIVFYRCKPKGVKUNWIMHEYRIIDNEAKSKPPGVYDACHNKSLRI
AcNAC4     GNS NRAATSGYKATC DKKP VLTSSGSOKVGVKKAIVFYRCKPKGVKUNWIMHEYRIIDNEAKSKPPGVYDACHNKSLRI

170          180          190          200          210          220          230          240
Consensus  DDWVLCRLYKKNP PRP IDHERD DSDMLRMPPTSS IPLCQHTPKLPPEKATSYG--S FUGNEHTI HEDMIGHDNTYISHLA
AaNAC1     DDWVLCRLYKKNP PRP IDHERD DSDMLRMPPTSS IPLCQHTPKLPPEKATSYG--S FUGNEHTI HEDMIGHDNTYISHLA
AcNAC1     DDWVLCRLYKKNP PRP IDHERD DSDMLRMPPTSS IPLCQHTPKLPPEKATSYG--S FUGNEHTI HEDMIGHDNTYISHLA
AaNAC3     DDWVLCRLYKKNP SRP IGHEDYSDMLRMPPTSS IPLCQHTPKLPPEKATSYG--S FUGNEHTI HEDMIGHDNTYISHLA
AcNAC3     DDWVLCRLYKKNP YSRP IGHEDYSDMLRMPPTSS IPLCQHTPKLPPEKATSYG--S FUGNEHTI HEDMIGHDNTYISHLA
AaNAC2     DDWVLCRLYKKNP PRP IDHERD DSDMLRMPPTSS IPLCQHTPKLPPEKATSYG--S FUGNEHTI HEDMIGHDNTYISHLA
AcNAC2     DDWVLCRLYKKNP PRP IDHERD DSDMLRMPPTSS IPLCQHTPKLPPEKATSYG--S FUGNEHTI HEDMIGHDNTYISHLA
AaNAC4     DDWVLCRLYKKNP PRP IDHERD DSDMLRMPPTSS IPLCQHTPKLPPEKATSYG--S FUGNEHTI HEDMIGHDNTYISHLA
AcNAC4     DDWVLCRLYKKNP PRP IDHERD DSDMLRMPPTSS IPLCQHTPKLPPEKATSYG--S FUGNEHTI HEDMIGHDNTYISHLA

250          260          270          280          290          300          310          320          330          332
Consensus  S----HLKRXLPPLYWDES ----XASKRHEGTSNIVASTXEDSSIASLLSXMPQTPTMXRIGDGGFXXPYSHPSLNWYS
AaNAC1     SKPOLPMKRALP SVYVDDVGEAGSSSKRHLHLDGNEGSSKTDENSMATLLSSLSOLPQTES-----MMDDVFRWYS
AcNAC1     SKPOLPMKRALP SVYVDDVGEAGSSSKRHLHLDGNEGSSKTDENSMATLLSSLSOLPQTES-----MMDDVFRWYS
AaNAC3     S----HLKRTLPPLYWANES ----EASKRHEGTSNIVASTXEDSSIASLLSQMFQTVTRRIGDGGFRQPHSHPSLNWYS
AcNAC3     S----HLKRTLPPLYWANES ----EASKRHEGTSNIVASTXEDSSIASLLSQMFQTVTRRIGDGGFRQPHSHPSLNWYS
AaNAC2     S----HLNRMLPPLYWDEG ----EASKRHEGTSNIVASTXEDSSIASLLRQMFQTVTRRIGDGGFRQPHSHPSLNWYS
AcNAC2     S----HLNRMLPPLYWDEG ----EASKRHEGTSNIVASTXEDSSIASLLRQMFQTVTRRIGDGGFRQPHSHPSLNWYS
AaNAC4     S----HLNRMLPPLYWDEG ----EASKRHEGTSNIVASTXEDSSIASLLRQMFQTVTRRIGDGGFRQPHSHPSLNWYS
AcNAC4     S----HLNRMLPPLYWDEG ----EASKRHEGTSNIVASTXEDSSIASLLRQMFQTVTRRIGDGGFRQPHSHPSLNWYS
```

B)



**Figure S7.**

(A) ClustalW alignment of the *Actinidia chinensis* (Ac) and *A. arguta* (Aa) NAC1-4 proteins.

(B) Unrooted Maximum Likelihood tree of selected *Arabidopsis thaliana* (Ooka et al., 2003), *A. arguta* (Aa) and *A. chinensis* (Ac) NACs. *A. arguta*: AaNAC1 (KF319046), AaNAC2 (KF319047), AaNAC3 (KF319048), AaNAC4 (KF319049), *A. chinensis*: AcNAC1 (KF319050), AcNAC2 (KF319051), AcNAC3 (KF319052), AcNAC4 (KF319053). The evolutionary history was based on the JTT matrix-based model (Jones et al., 1992). The percentage of trees in which the associated taxa clustered together is shown next to the branches. Initial tree(s) for the heuristic search were obtained automatically by applying Neighbor-Join and BioNJ algorithms to a matrix of pairwise distances estimated using a JTT model, and then selecting the topology with superior log likelihood value. The tree is drawn to scale, with branch lengths measured in the number of substitutions per site. All positions containing gaps and missing data were eliminated. There were a total of 158 positions in the final dataset. Evolutionary analyses were conducted in MEGA5 (Tamura et al., 2011).



A)

1 10 20 30 40 50 60 70 80 90 100 110  
AcEIL4 MEEICVD-----ISSDLVVDIIRCENLIDSDSEETIADLERRMWDKRIKLRKREKLAQAQAAKKEPKMSDQARRKKMSRAODGITKYLKLM  
AdEIL4 MEEICVD-----ISSDLVVDIIRCENLIDSDSEETIADLERRMWDKRIKLRKREKLAQAQAAKKEPKMSDQARRKKMSRAODGITKYLKLM  
AaEIL4 MEEICVD-----ISSDLVVDIIRCENLIDSDSEETIADLERRMWDKRIKLRKREKLAQAQAAKKEPKMSDQARRKKMSRAODGITKYLKLM  
AcEIL3 MDANCLD-----YNSDLVVDIIRCENLIDSDSEETIADLERRMWDKRIKLRKREKLAQAQAAKKEPKMSDQARRKKMSRAODGITKYLKLM  
AdEIL3 MDHFVLDANCLD-----DNSDLVVDIIRCENLIDSDSEETIADLERRMWDKRIKLRKREKLAQAQAAKKEPKMSDQARRKKMSRAODGITKYLKLM  
AcEIL1 MMTIDENCFCGDVDFSAPLGEGDVAFQTEQAVVDDSDSEEMVDLERRMWDKRIKLRKREKLAQAQAAKKEPKMSDQARRKKMSRAODGITKYLKLM  
AdEIL1 MMTIDENCFCGDVDFSAPLGEGDVAFQTEQAVVDDSDSEEMVDLERRMWDKRIKLRKREKLAQAQAAKKEPKMSDQARRKKMSRAODGITKYLKLM  
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120 130 140 150 160 170 180 190 200 210 220  
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AaEIL4 EVCKARGFVYGHPEKKGKPVSGSDNHRMWRKVKFKDKNGPAATIKYVABCLAK-VKERRIQKGSQSTIADTODHFGSITSSLVVQHCDPDRKYPLEKGVPEPWFVFGS  
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AcEIL1 EVCKARGFVYGHPEKKGKPVSGSDNHRMWRKVKFKDKNGPAATIKYVADNSIIPGKNGSGLVGFPHITADTODHFGSITSSLVVQHCDPDRKYPLEKGVPEPWFVFGS  
AdEIL1 EVCKARGFVYGHPEKKGKPVSGSDNHRMWRKVKFKDKNGPAATIKYVADNSIIPGKNGSGLVGFPHITADTODHFGSITSSLVVQHCDPDRKYPLEKGVPEPWFVFGS  
AcEIL2 EVCKARGFVYGHPEKKGKPVSGSDNHRMWRKVKFKDKNGPAATIKYVADNSIIPGKNGSGLVGFPHITADTODHFGSITSSLVVQHCDPDRKYPLEKGVPEPWFVFGS  
AdEIL2 EVCKARGFVYGHPEKKGKPVSGSDNHRMWRKVKFKDKNGPAATIKYVADNSIIPGKNGSGLVGFPHITADTODHFGSITSSLVVQHCDPDRKYPLEKGVPEPWFVFGS

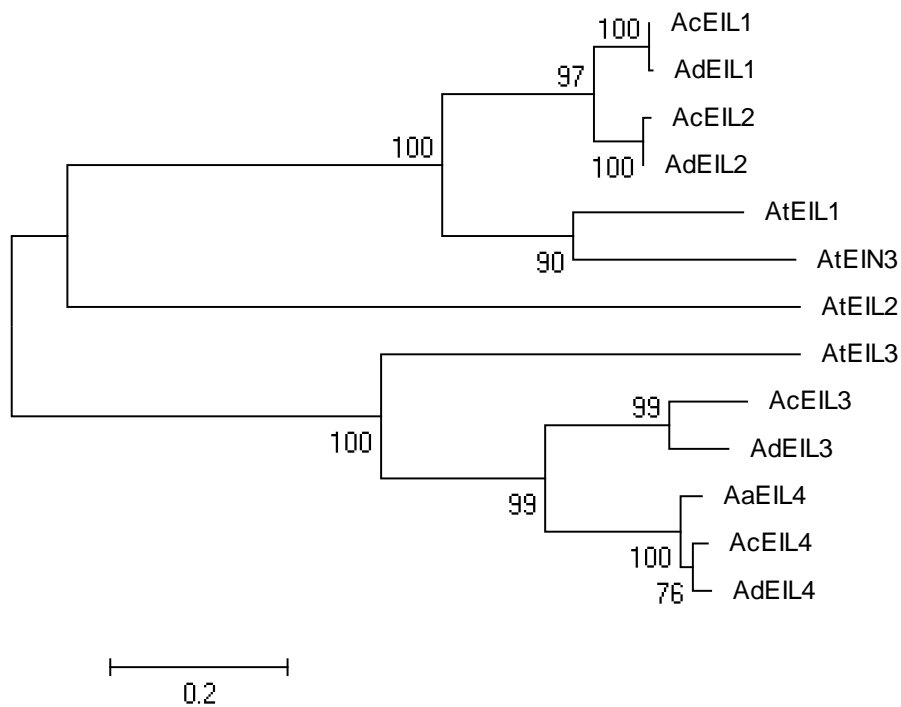
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AaEIL4 DEWVKTGLPKCSPPYKPHDLKKWKVGVLTAVTKHMSPDIAKRRLVRQSKCLQDKMTAKESAWLVLVLSREESLIRROSSDNGTSGTTEVPSGGGRRKKKPSVSSDS  
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AdEIL2 DEWVKTGLPKCSPPYKPHDLKKWKVGVLTAVTKHMSPDIAKRRLVQSKCLQDKMTAKESAWLVLVLSREESLIRROSSDNGTSGTTEVPSGGGRRKKKPSVSSDS

340 350 360 370 380 390 400 410 420 430 440  
AcEIL4 DYPVDGIDVGVSVSSKDDERDQLPDVEPSVQ-----RNSTARLQDKERREEQPRRKRPRVRSPTNQAAFPYSRHLDHDEPINTLPDINLHVDLSHMHGSOHNNANDE  
AdEIL4 DYPVDGIDVGVSVSSKDDERDQLPDVEPSVQ-----RNSTARLQDKERREEQPRRKRPRVRSPTNQAAFPYSRHLDHDEPINTLPDINLHVDLSHMHGSOHNNANDE  
AaEIL4 DYPVDGIDVGVSVSSKDDERDQLPDVEPSVQ-----RNSTARLQDKERREEQPRRKRPRVRSPTNQAAFPYSRHLDHDEPINTLPDINLHVDLSHMHGSOHNNANDE  
AcEIL3 DYPVDGIDVGVSVSSKRRTDQLLDVEPSVQ-----RNTKPHSLRTERREEQPRRKRPRVRSPTNQAAFPYSRHLDHDEPINTLPDINLHVDLSHMHGSOHNNANDE  
AdEIL3 DYPVDGIDVGVSVSSKRRTDQLLDVEPSVQ-----RNTKPHSLRTERREEQPRRKRPRVRSPTNQAAFPYSRHLDHDEPINTLPDINLHVDLSHMHGSOHNNANDE  
AcEIL1 DYPVDGIDVGVSVSSKRRTDQLLDVEPSVQ-----RNTKPHSLRTERREEQPRRKRPRVRSPTNQAAFPYSRHLDHDEPINTLPDINLHVDLSHMHGSOHNNANDE  
AdEIL1 DYPVDGIDVGVSVSSKRRTDQLLDVEPSVQ-----RNTKPHSLRTERREEQPRRKRPRVRSPTNQAAFPYSRHLDHDEPINTLPDINLHVDLSHMHGSOHNNANDE  
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450 460 470 480 490 500 510 520 530 540 550 560  
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AcEIL2 FVLTBSFRNIG--TKPIVIGDFPFWC KPAAPSVDHRSFDSGLGWDKMI SELVSYDANI QNRCSNPMNISTSSL-----NQLSQSLDQPNLCCQDNNYIHG  
AdEIL2 FVLTBSFRNIG--TKPIVIGDFPFWC KPAAPSVDHRSFDSGLGWDKMI SELVSYDANI QNRCSNPMNISTSSL-----NQLSQSLDQPNLCCQDNNYIHG

570 580 590 600 610 620 630 640 650 660 667  
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AdEIL4 CQQTQMAFTEPWRDREVDGIVNASALRRNDIPNPGGDMIDYIKGTQONODRPMVNHGSPNSLSLDY GGLNSPFNLEIHGTSSTDTSDIDFLD DDTIQCFCA  
AaEIL4 CQQTQMAFTEPWRDREVDGIVNASALRRNDIPNPGGDMIDYIKGTQONODRPMVNHGSPNSLSLDY GGLNSPFNLEIHGTSSTDTSDIDFLD DDTIQCFCA  
AcEIL3 CQQTQMAFTEPWRDREVDGIVNASALRRNDIFG--DMPP--PNEEAPVANFLGSPNSLSLDY-----HLGTDALGSDTADVDSFDDNIMBMLCA  
AdEIL3 CQQTQMAFTEPWRDREVDGIVNASALRRNDIFG--DMPP--PNEEAPVANFLGSPNSLSLDY-----HLGTDALGSDTADVDSFDDNIMBMLCA  
AcEIL1 CFVEG-HVHECANLQNSMFSQOENR D-----RQVNSQEPANPNDFPIMGSPDYLFQMDY P-----EHLPAVAR-DITSKPDES IWF  
AdEIL1 CFVEG-HVHECANLQNSMFSQOENR D-----RQVNSQEPANPNDFPIMGSPDYLFQMDY P-----EHLPAVAR-DITSKPDES IWF  
AcEIL2 CFVMEKNTCCCANLQNSMFSQOENR D-----RQVNSQEPANPNDFPIMGSPDYLFQMDY P-----EHLPAVAR-DITSKPDES IWF  
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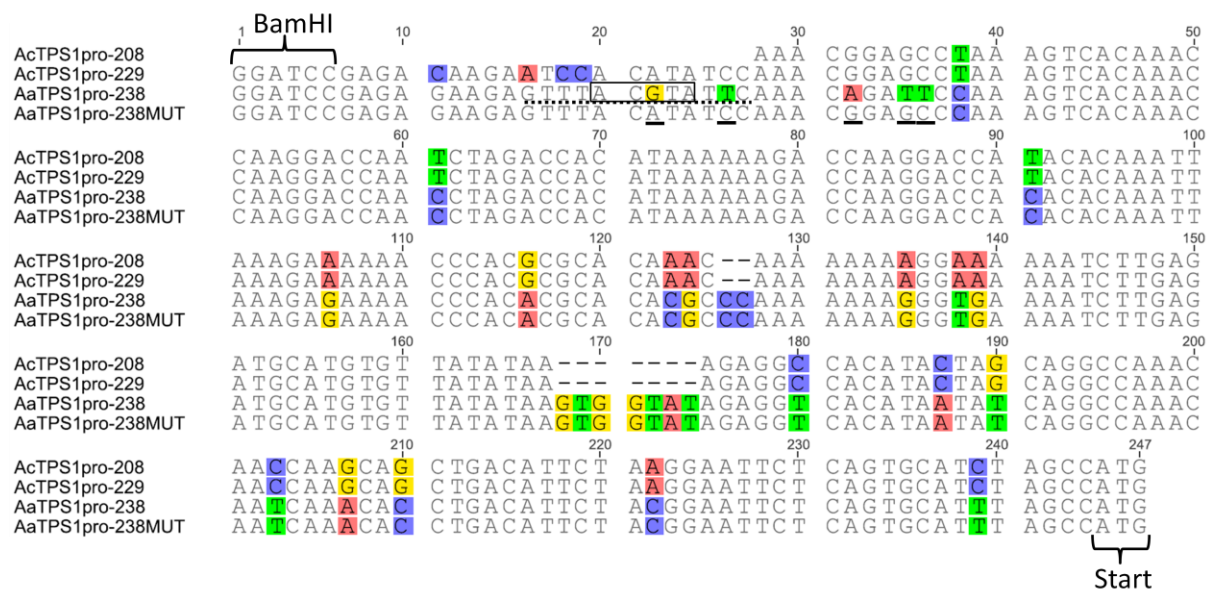
B)



**Figure S8.**

(A) ClustalW alignment of the *Actinidia deliciosa* (Ad), *A. chinensis* (Ac) and *A. arguta* (Aa) *EIL1-4* proteins.

(B) Unrooted Maximum Likelihood tree of *A. deliciosa* (Ad), *A. arguta* (Aa) and *A. chinensis* (Ac) EIN3-like (EIL) transcription factors compared to *Arabidopsis thaliana*. Arabidopsis (At) EIN3 (GenBank AAC49749), EIL1 (AAC49746), EIL2 (AAC49747) and EIL3 (AAC49748), *A. deliciosa* (Yin et al., 2010): AdEIL1 (EU170633), AdEIL2 (EU887511), AdEIL3 (EU887512), AdEIL4 (EU887513), *A. chinensis*: AcEIL1 (KF319041), AcEIL2 (KF319042), AcEIL3 (KF319043), AcEIL4 (KF319044), *A. arguta*: AaEIL4 (KF319045). The tree was generated as described in Fig S7. There were a total of 239 positions in the final dataset.



**Figure S9.** Sequence alignment of minimal Aa *TPS1*<sub>pro</sub> and Ac *TPS1*<sub>pro</sub> promoter fragments. The conserved NAC binding core ACGTA is boxed in Aa *TPS1*<sub>pro</sub>-238 and substitutions in *pro*-238Mut are underlined. The NAC 12 bp (GTTTACGTATTC) sequence fused to Ac *TPS1*<sub>pro</sub>-208 (see Fig 8E) is highlighted with a dotted line. The translational ATG start codon and *Bam*HI cloning site are highlighted.

**Table S1.** Characteristics of *Actinidia arguta* ‘Hortgem Tahi’ and *A. chinensis* ‘Hort16A’ ripening fruit. SSC: soluble solids content. nd: not determined

<b>Species</b>	<b>ripening stage</b>	<b>firmness (N)</b>	<b>fruit weight (g)</b>	<b>SSC (°Brix)</b>
<i>A. arguta</i>	a0	7.80 ± 0.33	13.46 ± 0.57	nd
<i>A. arguta</i>	a1	2.50 ± 0.02	14.74 ± 0.44	11.74 ± 0.19
<i>A. arguta</i>	a2	1.72 ± 0.02	15.50 ± 0.46	12.13 ± 0.30
<i>A. arguta</i>	a3	1.26 ± 0.01	14.10 ± 0.41	12.42 ± 0.22
<i>A. arguta</i>	a4	0.83 ± 0.02	13.53 ± 0.54	13.76 ± 0.81
<i>A. arguta</i>	a5	0.63 ± 0.01	13.29 ± 0.44	13.20 ± 0.67
<i>A. chinensis</i>	c0	31.1 ± 3.0	nd	16.66 ± 0.42
<i>A. chinensis</i>	c1	8.4 ± 0.5		19.11 ± 0.22
<i>A. chinensis</i>	c2	6.5 ± 0.3		18.83 ± 0.26
<i>A. chinensis</i>	c3	3.1 ± 0.2		19.33 ± 0.34
<i>A. chinensis</i>	c4	2.6 ± 0.1		18.89 ± 0.34

**Table S2.** Cloning and PCR primers

<b>Primer</b>	<b>Sequence</b>
<b>A) Transient terpene synthase and MEP pathway gene over-expression of pHEX2 vector in Nicotiana benthamiana</b>	
AacTPS1_F1	CACCATGTCTCTTAGTGTGGTTTTGG
AaTPS1_R1	CTAAGCTTTCTCCTCAAAGGGAATGGGACTGATTAGTAG
AcTPS1_R1	TTAAAAAAGGGGAATGGGATTGATTAGTAGTACTGACTCG
AdHDRF1	GGGGACAAGTTTGACAAAAAGCAGGCTATGATGATTCTCTGCAATTCTG
AdHRR1	GGGGACCACTTTGTACAAGAAAGCTGGGTCTAAGCCAATTGCAGGGC
AcDXSF	CACCATGGCTGCTAGTGTGTTCTC
AcDXSR	CAAGGATTATTCTTAGTTGAAAGGA
AaDXRF1	GGGGACAAGTTTGACAAAAAGCAGGCTATGGCTCTAAATTTGCTATCCC
AaDXRR1	GGGGACCACTTTGTACAAGAAAGCTGGGTTCATACAGGAACAGGACTC
AaIDIF1	GGGGACAAGTTTGACAAAAAGCAGGCTATGTCCTCTCTCTCC
AaIDIR1	GGGGACCACTTTGTACAAGAAAGCTGGGTTAAGTCAATTTGTGAATGGTTTTTCAT
<b>B) NAC and EIL plant over-expression in pHEX2</b>	
pH_AacNAC1F	AAAAAGCAGGCTCCATGGAGAGCACGGATTCCG
pH_AacNAC1R	AGAAAGCTGGGTCTAGGAGTACCATCGAAAG
pH_AacNAC2F	AAAAAGCAGGCTCCATGGAGAGCCCGATTCCG
pH_AacNAC2R	AGAAAGCTGGGTAGGGTACCAATTCAGCTAG
pH_AacNAC3F	AAAAAGCAGGCTCCATGGAGAGCACGGATTCA
pH_AacNAC3R	AGAAAGCTGGGTCTAAGAGTACCAATTCAGCTAG
pH_AacNAC4F	AAAAAGCAGGCTCCATGGAGAAGCTCAACGTTG
pH_AacNAC4R	AGAAAGCTGGGTCTAAGTTTTCTTTAAAGGATGAG
pH_AcEIL1F	AAAAAGCAGGCTGGATGATGATATTTGATGAAATGGGG
pH_AcEIL1R	AGAAAGCTGGGTCTAGAACCAATCGAGCC
pH_AcEIL2F	AAAAAGCAGGCTGGATGATAATGTTGACGAGATGGG
pH_AcEIL2R	AGAAAGCTGGGTCTAGAACCATACTGAGCC
pH_AcEIL3F	AAAAAGCAGGCTGGATGGATGCCAATGGGTTG
pH_AcEIL3R	AGAAAGCTGGGTCTATGCCCAAGGTATTC
pH_AacEIL4F	AAAAAGCAGGCTGGATGGAAGAGATTGGAGTTGATAT
pH_AacEIL4R	AGAAAGCTGGGTTATGCACCAAAGCATTGGATC
<b>C) Real time PCR analysis</b>	
<b>TPS genes</b>	
TPSRTF1 (Fig. 2)	CGTTGAGAGGTGGGATCTGAGTGAA
TPSRTR1 (Fig. 2)	GACACCTTGCTCCTTAGGGGTGCA
TPSUF (Fig. S9)	TTGCCATATTGGTGAGTCATGCCTT
TPSUR ((Fig. S9)	TGTGGGACTCAAATTAGGTCTCTCTCA
<b>MEP genes</b>	
DXS_F1	GCCTCCCTGTTTGAAGAACTTGGAT
DXS_R1	CAGGCATGGCCTTGACCTTCTT
DXR_F1	TGCAAGCTTGGGTCACTGACATTT
DXR_R1	CGGCTTTCTCATTTCAGCACTAA
MCT_F1	CACTCGCATCTGCAGCTAACCAGTT
MCT_R1	TCCACAGCCTTGGCAGAGCAA
CMK_F1	TATTGTCCACGGCTTGACAGCAA
CMK_R1	CAATGATTTGGAACCTCCTGCATTT
MDS_F1	TCGAGCCAGGTTACCCTCTCATCAT

MDS_R1	GGCGCCAAAATTGCATCAA
HDS_F1	TGGAGACCACCAACATACCCAA
HDS_R1	TGCCCGGTGTTTCTATTGCCAT
HDS_F2	CCGTCTTTGTATTCTCATCTGCA
HDS_R2	TAGATGGTCTTGGAGATGGCATTCT
HDR_F1	CACAATAGGGGTAACATCTGGTGCC
HDR_R1	CAATTGCAGGGCTTCTTCTCGTTTA
HDR_F2	ATGATGTTGTGGTTTTGCCTGCAT
HDR_R2	CCAAACCTTAGAAACCCATGGACAA
IDI_F1	CGAACCTGATGAAGTTGCTGACAT
IDI_R1	TGAACCATGGGGACAGCTTCAAA
ssuGDPSII_F1	CTTCGAGCCCATGCACCACCTA
ssuGDPSII_R1	CTCACACGCGGCGACGCATA
ssuGDPSII_F2	CCCGTCTTCCATGGCCGTAT
ssuGDPSII_R2	CGGAACTAGCCTTGTGGACCATGTA

***NAC genes***

AcNAC1F	AAGAGCAACCTCCCAAGACCAA
AaNAC1F	AAGAGCAACCACCCACGACCAT
NAC1R	GAAGGTGGCATTCTTGAAGCAT
NAC2F	CCGGTGGCGATCATAGCTGAAAT
NAC2R	CCGGTTCCTCGGACTGAAAAAT
NAC3F	CAAACCCAAAACCTCCACCAA
NAC3R	GCTGTGACTGAGCATTTCGTCGAA
NAC4F	TAGGACTGATTGGATCATGCACGAA
NAC4R	CGGCAAAGAACCCAATTCTCCATA

***EIL genes***

AcEIL1F	GAGAATCGGTTTGATCAGCGCAA
AcEIL1R	CCGAACATCAAGGGGAAGTTATCATT
AcEIL2F	GGGCAAAAATGATCAACGAGCTT
AcEIL2R	GAAGAACTGAGATGCTCATGGGATTT
AcEIL3F	ACACAGAACGTATTTGTTGGTGGCAT
AcEIL3R	TGAGATTCATGATGCAACTCGGAGTT
AcEIL4F	ACCGGATGTGGAACCATCTAATCAA
AcEIL4R	CTCTCGTCGCTCTTTGTCTTGCAA
AaEIL4F	AATCAACCACCATCTAATCAACTCGA
AaEIL4R	AGGGCTTGATCTTACTCGTGATCTTTTT

***Reference***

AdEF1F	GCACTGTCATTGATGCTCCT
AdEF1R	CCAGCTTCAAAACCACCAT

***D) Bacterial protein expression of Ac- and AaTPS1 in the pET200 vector***

p200_AcTPS1F	CACCGAAAACCTGTATTTTCAGGGGAATTGCGGAGCCTAGCCAAACC
p200_AcTPS1R	TTAAAAAAGGGGAATGGGATTGATTAGTAGTACTTGACTCG
p200_AaTPS1F	CACCAGGCGATCCGCGTTCTACCAACCTTC
p200_AaTPS1R	CTAAGCTTTCTCCTCAAAGGGAATGGGACTGATTAGTAG

***E) Promoter constructs of pGreenII 0800-LUC***

F1_717	AAACATAGTGCATATTGTGCGTCTGA
F2_757	AATCGATCGTAACATTCACACAACCG
R1_131	GGTCCGAAATCGTGATCGAA

R2_101	CGCATATGTACCCTTCGATGATTGG
AaproF	TTAAGGATCCTTTACTAGATTTAAAAATTAATTTTAACATA
AaproR	TTAACCATGGCTAAATGCACTGAGAATTC
AcproF	TTAAGGATCCCTCCTAAAATAGAGTTACAAAAA
AcproR	TTAACCATGGCTAGATGCACTGAG
F-238	TTAAGGATCCGAGAGAAGAGTTTACGTATTCAA
F-229	TTAAGGATCCGAGACAAGAATCCACATATCC
F-238mut	TTAAGGATCCGAGAGAAGAGTTTACATATCCAAACGGAGCCCAAAGTCACAAACCAA
F-600	AAAAGGATCCTAAGGAAAATAAAAYCAAACACTA
208 (I)F	TTAAGGATCCGTTTACGTATTCAAACGGAGCCTAAAGTCACA
208 (II)F	TTAAGGATCCGTTTACGTATTCTGTTTACGTATTCTGTTTACGTATTCAAACGGAGCCTAAAGTCACA
208 (III)F	TTAAGGATCCGTTTACGTATTCTGTTTACGTATTCTGTTTACGTATTCTGTTTACGTATTCTGTTTACGTATTCAAACGGAGCCTAAAGTCACA

***F) Bacterial protein expression of the NAC DNA binding domains in pET300***

AaNAC1DBF	AAAAAGCAGGCTCCATGGAGAGCACGGATTTCG
AaNAC1DBR	AGAAAGCTGGGTTTCATGGTCGTGGGTGGTT
AaNAC2DBF	AAAAAGCAGGCTCCATGGAGAGCCCGGATTTCG
AaNAC2DBR	AGAAAGCTGGGTTTCAGGCTCTTGGTGGGT
AaNAC3DBF	AAAAAGCAGGCTCCATGGAGAGCACGGATTCA
AaNAC3DBR	AGAAAGCTGGGTTTCAGGCTTGTGATGGGTG
AaNAC4DBF	AAAAAGCAGGCTCCATGGAGAAGCTCAACGTTG
AaNAC4DBR	AGAAAGCTGGGTTCAATTTTAGTACTTCTTTCTCAAAAATA

**Table S3.** Ripe fruit headspace terpene compounds in fourteen kiwifruit species. Headspace volatile levels were determined by dynamic headspace sampling of pulped fruit for 20 minutes and compounds were identified by GC-MS.

Compound (ng . g <sup>-1</sup> . h <sup>-1</sup> )	DB WAX Retention index (s)	Species ( # accessions)																														
		<i>A. arguta</i> AVERAGE (n=15)		<i>A. chinensis</i> AVERAGE (n=8)		<i>A. chinensis</i> AVERAGE (n=2)		<i>A. deliciosa</i> AVERAGE (n=3)		<i>A. esculenta</i> AVERAGE (n=5)		<i>A. glaucophylla</i> AVERAGE (n=1)		<i>A. indicochinensis</i> AVERAGE (n=2)		<i>A. latifolia</i> AVERAGE (n=3)		<i>A. macrocarpa</i> AVERAGE (n=3)		<i>A. melanandra</i> AVERAGE (n=3)		<i>A. polygama</i> AVERAGE (n=5)		<i>A. purpurea</i> AVERAGE (n=4)		<i>A. ujo</i> AVERAGE (n=2)		<i>A. zircosa</i> AVERAGE (n=1)				
		MAX	AVERAGE	MAX	AVERAGE	MAX	AVERAGE	MAX	AVERAGE	MAX	AVERAGE	MAX	AVERAGE	MAX	AVERAGE	MAX	AVERAGE	MAX	AVERAGE	MAX	AVERAGE	MAX	AVERAGE	MAX	AVERAGE	MAX	AVERAGE	MAX	AVERAGE			
unknown 1	1021	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0		
pinene, alpha	1026	245.7	3248.5	0.6	7.2	68.2	157.4	0.0	0.0	33.9	63.8	144.4	159.5	2.2	6.5	0.0	0.0	0.0	0.0	24.1	46.5	38.6	87.6	13.9	55.4	27.7	55.4	0.0	0.0	0.0	0.0	
thujene, alpha	1033	5.2	42.9	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	2.1	4.5	30.4	191.8	55.3	213.3	110.6	213.3	0.0	0.0	0.0	0.0	
fenchene, alpha	1051	0.0	0.6	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
camphene	1063	2.4	26.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.6	1.7	0.6	2.5	1.4	5.4	2.7	5.4	0.0	0.0	0.0	0.0	
pinene, beta	1107	137.7	1654.2	2.4	83.3	11.9	29.2	0.0	0.0	27.1	84.2	11.1	16.5	0.0	0.0	0.0	0.0	0.0	0.0	9.0	20.1	20.0	48.1	14.4	28.9	24.9	28.9	3.9	7.8	0.0	0.0	
terpinene, beta or sabinene	1121	151.3	2022.5	0.4	5.1	0.0	0.0	0.0	0.0	2.3	13.9	156.0	168.8	0.0	0.0	0.0	0.0	0.0	0.0	2.1	5.0	66.8	200.0	1.0	4.0	2.0	4.0	0.0	0.0	0.0	0.0	
menthene, p	1132	0.0	0.0	5.3	82.7	2.9	7.2	2.6	15.5	2.0	11.9	0.0	0.0	6.8	20.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
terpinene, alpha	1172	63.8	1084.6	0.2	5.3	5.7	22.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.7	2.1	0.4	1.6	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
myrcene, beta	1169	879.8	7596.4	0.0	0.0	16.7	34.7	0.0	0.0	6.9	41.1	22.3	35.3	0.0	0.0	0.0	0.0	0.0	0.0	97.4	187.6	155.7	535.2	37.4	95.4	27.2	51.5	47.7	95.4	0.0	0.0	
limonene	1198	289.8	3331.0	1.0	18.7	223.6	558.9	0.0	0.0	18.8	64.2	72.6	93.8	3.3	9.8	0.0	0.0	0.0	0.0	80.4	144.0	58.5	168.6	17.7	67.7	33.9	67.7	1.6	3.2	0.0	0.0	
phellandrene, beta	1215	26.6	455.7	0.0	0.0	2.4	9.5	0.0	0.0	0.0	0.0	2.0	2.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	8.1	44.9	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
eucalyptol	1216	427.9	7529.0	66.7	333.2	6.4	25.6	0.0	0.0	0.0	0.0	0.0	0.0	19.1	57.2	0.0	0.0	0.0	0.0	0.0	0.0	63.2	144.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
unknown 2	1217	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	68.5	271.3	135.7	271.3	1.4	2.7	0.0	0.0	
ocimene, (Z)-beta	1230	9.1	107.2	0.0	0.0	1.7	6.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.2	8.4	36.7	146.7	0.0	0.0	73.4	146.7	0.0	0.0		
terpinene, gamma	1250	146.8	2455.1	0.6	12.0	2.7	9.2	0.0	0.0	0.0	0.0	0.0	0.0	15.5	19.2	0.0	0.0	0.0	0.0	0.2	0.5	10.5	25.4	0.4	1.5	0.8	1.5	0.0	0.0	0.0	0.0	
ocimene, (E)-beta	1230	9.1	107.2	0.0	0.0	1.7	6.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.2	8.4	36.7	146.7	0.0	0.0	73.4	146.7	0.0	0.0		
unknown 3	1267	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
cymene, p	1268	163.9	2729.0	17.4	260.0	10.5	26.2	1.0	6.0	0.2	0.9	73.7	101.0	0.0	0.0	0.0	0.0	0.0	0.0	4.3	13.0	9.5	25.9	1.9	7.5	3.8	7.5	0.0	0.0	0.0	0.0	
dimethylstyrene	1278	161.9	1755.7	0.2	9.1	253.2	571.7	0.0	0.0	24.3	120.8	20.6	29.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	4.8	21.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
terpinolene	1285	1132.8	7546.9	0.7	13.0	160.7	537.5	29.4	176.2	253.4	869.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.9	489.1	1193.2	0.0	0.0	174.5	697.8	348.9	697.8	0.0	0.0	0.0	0.0
terpinolene, iso	1286	45.7	391.5	0.2	3.2	2300.1	5208.5	0.0	0.0	38.3	134.8	12.9	16.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.9	3.3	2.5	9.8	4.9	9.8	0.0	0.0	
unknown 4	1408	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	6.7	37.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
perillen	1409	1.4	25.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
unknown 5	1414	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.8	2.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
mentha-1,3,8-triene, p	1399	12.1	128.5	0.0	0.0	1.9	7.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.5	1.5	0.0	0.0	0.3	1.0	0.5	1.0	0.0	0.0	0.0	0.0	
unknown 6	1461	0.6	11.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
linalool	1538	0.4	5.7	0.0	0.0	12.9	35.6	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	2.6	10.3	0.0	0.0	16.0	51.1	0.0	0.0	0.0	0.0	0.0	0.0	
linalool acetate	1538	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.4	1.5	0.0	0.0	1.4	9.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
unknown 8	1560	0.0	0.0	0.0	0.0	55.8	193.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
unknown 9	1582	1.4	24.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.5	4.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
terpine-4-ol	1600	74.5	1341.9	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
Unknown 10	1620	1.3	23.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	2.3	6.8	0.0	0.0	0.0	0.0	0.0	0.0	
terpineol, alpha	1695	124.0	1988.3	0.0	0.0	5.4	17.9	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	31.4	118.9	0.0	0.0	0.0	0.0	0.0	0.0	
citral	1704	1.2	15.3	0.0	0.0	19.6	52.9	0.0	0.0	0.0	0.0	10.8	16.3	0.0	0.0	0.0	0.0	0.0	0.0	3.3	8.7	0.1	1.0	1.1	4.4	2.2	4.4	0.0	0.0	0.0	0.0	
geranylacetone	1835	0.0	0.0	16.3	192.4	0.0	0.0	8.0	21.5	0.0	0.0	0.0	0.0	1.7	5.1	2.8	8.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
cymen-8-ol, p	1851	1.6	24.1	0.0	0.0	42.1	103.5	0.0	0.0	0.0	0.0	1.9	3.8	0.0	0.0	0.0	0.0	0.0	0.0	3.3	9.1	0.5	3.2	1.9	7.4	3.7	7.4	0.0	0.0	0.0	0.0	
geranylacetone	1855	3.0	15.7	3.3	125.0	3.5	9.0	0.0	0.0	2.5	13.9	0.0	0.0	11.5	29.8	2.9	5.7	2.7	8.9	0.0	0.0	5.8	18.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	

Key: >10 ng . g<sup>-1</sup> . h<sup>-1</sup>  
>100 ng . g<sup>-1</sup> . h<sup>-1</sup>



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