

Supplementary material for the manuscript:

The ambiguous ripening nature of the fig (*Ficus carica* L.) fruit: a gene-expression study of potential ripening regulators and ethylene-related genes

Zohar E. Freiman^{1,2}, Yogev Rosiansky^{1,2}, Rajeswari Dasmohapatra¹, Itzhak Kamara¹ and Moshe A. Flaishman^{1,*}

¹ Institute of Plant Sciences, Agricultural Research Organization, P.O. Box 6, Bet-Dagan 50250, Israel

² The Robert H. Smith Institute of Plant Sciences and Genetics in Agriculture, Faculty of Agriculture, Food and Environment, The Hebrew University of Jerusalem, P.O. Box 12, Rehovot 76100, Israel

* To whom correspondence should be addressed. E-mail: vhmoshea@agri.gov.il

Table S1. Fig genes homologous to *MADS-box*, ethylene-synthesis and ethylene-signal-transduction genes subjected to gene-expression analysis. Homologous genes from *Arabidopsis*, tomato and apple are noted.

Gene name (sequence ID of cDNA isolates)	Arabidopsis homologue (Protein sequence ID, NCBI)	Tomato homologue (Protein sequence ID, NCBI)	Apple homologue (Protein sequence ID, NCBI)
<i>FcMADS2</i> (KJ506153)	AP1 (NP_177074.1)	MADS-MC (NP_001234665.1)	MADS5 (ABG85297.1)
<i>FcMADS3</i> (KJ506154)	FUL, AGL8 (NP_568929.1)	FUL1 (AAP83379.1)	MADS2.1 (ABB22023.1)
<i>FcMADS4</i> (KJ506155)	SVP (BAD43004.1)	JOINTLESS-like (XP_004250145.1)	JOINTLESS (ABD66219.2)
<i>FcMADS5</i> (KJ506156)	SEP3 (NP_850953.1)	AGL31-Like (XP_004239235.1)	MADS9 (CAA04920.1)
<i>FcMADS6</i> (KJ506157)	AGL2 (AAA32732.1)	MADS1 (NP_001234380.1)	MADS3 (AAD51422.1)
<i>FcMADS8</i> (KJ506159)	SEP1 (NP_568322.1)	RIN (NP_001234670.1)	MADS4 (AAD51423.1)
<i>FcACSIL</i>	ACS6 (NP_192867.1)	ACS1B (AAF97615.1)	ACS-5B (BAA92351.1)
<i>FcACS2</i> (DQ269493.1)	ACS1 (NP_191710.1)	ACS1A (AAF97614.1)	ACS-5B (BAA92351.1)
<i>FcACS3</i> (DQ269494.1)	ACS8 (NP_195491.1)	ACS8 (AAQ13597.1)	ACS-1 (AAA73941.1)
<i>FcACS4</i>	ACS8 (NP_195491.1)	ACS3 (AAA78789.1)	ACS-1 (AAA73941.1)
<i>FcACOL</i> (AB307720.1)	ACO4 (NP_171994.1)	ACO1 (CAA41212.1)	ACO1 (AAC36461.1)
<i>FcACO2</i>	ACO5 (NP_565154.1)	ACO5-Like (XP_004242046.1)	ACO2 (AAB94031.1)
<i>FcACO3</i>	ACO1 (NP_179549.1)	ACO5 (NP_001234037.1)	ACO2 (AAB94031.1)
<i>FcEOL1</i>	ETO1 (AAT01656)	ETO1-Like	Low homology

		(XP_004247013.1)	
<i>FcEOL2</i>	ETO1-Like 1 (NP_192177.1)	EOL1 (AAZ08351.1)	Low homology
<i>FcETR1</i>	ETR1 (NP_176808.3)	ETR1 (NP_001234149.1)	ETR1b (AAW69924.1)
<i>FcETR2</i>	ETR2 (NP_188956.1)	ETR4 (NP_001234205.1)	ETR2 (ABI58286.1)
<i>FcEIN4</i>	EIN4 (NP_187108.1)	ETR5 (NP_001234212.1)	ETR2 (ABI58286.1)
<i>FcERS1</i>	ERS1 (NP_181626.1)	Nr (NP_001233894.1)	ERS2 (BAE97296.1)
<i>FcCTR1</i>	CTR1 (NP_195993.1)	CTR3 (NP_001234454.1)	CTR1 (ABI58288.1)
<i>FcCTR2</i>	CTR1 (NP_195993.1)	TCTR1 (NP_001234330.1)	CTR1 variant (ABI58289.1)
<i>FcCTR3</i>	CTR1 (NP_195993.1)	CTR3 (NP_001234454.1)	CTR1 (ABI58288.1)
<i>FcEDR1</i>	EDR1 (ABR45974.1)	CTR2 (NP_001234768.1)	CTR2 (ABI33217.1)
<i>FcEIN2</i>	EIN2 (NP_195948.1)	EIN2 (NP_001234518.1)	EIN2 (ABI33219.1)
<i>FcEBF1</i>	EIN3-binding F-box protein 1 (NP_565597.1)	EIN3-binding F-box protein 1 (NP_001234858.1)	EIN3 binding F-box 1 (AFT92041.1)
<i>FcEIL1</i>	EIL3 (NP_177514.1)	EIL3 (NP_001234546.1)	EIL1 (ADE41153.1)
<i>FcEIL2</i>	EIN3 (NP_188713.1)	EIL3 (NP_001234546.1)	EIL2 (ADE41154.1)
<i>FcEIL3</i>	EIN3 (NP_188713.1)	EIL3 (NP_001234546.1)	EIL3 (ADE41155.1)
<i>FcERF9168</i>	ERF071 (NP_182274.1)	ethylene response factor 2 (NP_001234308.1)	ERF071-like (XP_008386662.1)
<i>FcERF12185</i>	ethylene-responsive transcription factor TINY (NP_197953.1)	dehydration-responsive element-binding protein 3- like (XP_004241746.1)	AP2 domain class transcription factor (ADE41141.1)
<i>FcERF9211</i>	ERF118 (NP_177022.1)	ERF118-like isoform 1 (XP_004238935.1)	ethylene-responsive transcription factor CRF6- like (NP_001280962.1)
<i>FcERF9297</i>	ERF118 (NP_177022.1)	ERF118-like isoform 1 (XP_004238935.1)	ERF118-like (XP_008368213.1)
<i>FcERF8231</i>	ethylene-responsive	RAP2.6-like protein	AP2 domain class

	transcription factor ABR1 (NP_201280.1)	(ABY57638.1)	transcription factor (ADE41125.1)
<i>FcERF5086</i>	ERF061 (NP_176620.1)	ERF061-Like (XP_004245520.1)	ERF061 (XP_008341502.1)
<i>FcERF9572</i>	ERF109 (NP_195167.1)	RAP2.6-like protein (ABY57638.1)	AP2 domain class transcription factor (ADE41151.1)
<i>FcERF9690</i>	ethylene responsive element binding factor 1 (BAA32418.1)	ethylene-responsive transcription factor 1-like (XP_004235186.1)	ethylene-responsive transcription factor 2-like (XP_008352246.1)
<i>FcERF9109</i>	ERF025 (NP_200015.1)	ERF025-like (XP_004240742.1)	ERF027-like (XP_008381395.1)
<i>FcERF11513</i>	dehydration-responsive element-binding protein 1F (NP_172721.1)	dehydration-responsive element-binding protein 1C-like (XP_004234351.1)	AP2 domain class transcription factor (ADE41100.1)
<i>FcERF11632</i>	dehydration-responsive element-binding protein 1D (NP_200012.1)	dehydration-responsive element-binding protein 1C-like (XP_004234351.1)	AP2 domain class transcription factor (ADE41100.1)
<i>FcERF12049</i>	ethylene-responsive transcription factor (NP_188965.1)	ethylene responsive element binding protein (NP_001234695.1)	AP2 domain class transcription factor (ADE41122.1)
<i>FcERF8090</i>	ethylene-responsive transcription factor CRF4 (NP_194524.2)	pathogenesis-related genes transcriptional activator PTI6 (NP_001233991.1)	pathogenesis-related genes transcriptional activator PTI6-like (XP_008384656.1)
<i>FcERF10238</i>	ethylene-responsive transcription factor 9 (NP_199234.1)	ethylene response factor 3 (AAO34705.1)	ethylene-responsive transcription factor 4 (XP_008371404.1)
<i>FcERF10715</i>	ethylene-responsive transcription factor 9 (NP_199234.1)	ethylene-responsive transcription factor 4-like (XP_004243371.1)	ethylene-responsive transcription factor 4-like (XP_008358676.1)
<i>FcERF998</i>	ethylene-responsive transcription factor RAP2- 2 (NP_850582.1)	transcription factor JERF1 (NP_001234513.1)	ethylene-responsive transcription factor RAP2- 12-like (NP_001280975.1)
<i>FcERF14722</i>	dehydration-responsive	dehydration-responsive	putative dehydration-

	element-binding protein 2A (NP_001031837.1)	element-binding protein (NP_001234689.1)	responsive element-binding protein 2H (XP_008362249.1)
<i>FcERF2127</i>	ERF060 (NP_195688.1)	ethylene-responsive transcription factor RAP2-4-like isoform 1 (XP_004237407.1)	ERF060 (NP_001288047.1)
<i>FcERF9816</i>	ethylene-responsive transcription factor RAP2-1 (NP_564496.1)	ERF010-like (XP_004238136.1)	ERF011-like (XP_008378018.1)
<i>FcERF14757</i>	ERF053 (NP_179685.1)	ERF054-like (XP_004243339.1)	ERF054-like (XP_008342928.1)
<i>FcERF8610</i>	putative AP2 domain transcription factor (AAM65663.1)	AP2-like ethylene-responsive transcription factor At2g41710-like (XP_004231573.1)	AP2-like ethylene-responsive transcription factor At2g41710 (XP_008387305.1)
<i>FcERF3350</i>	ERF106 (AAK97736.1)	ethylene response factor 4 (NP_001234313.1)	ERF106-like (XP_008376371.1)
<i>FcERF9006</i>	ERF114 (NP_200995.1)	ERF115-like (XP_004235847.1)	ERF114-like (XP_008356833.1)
<i>FcERF1362</i>	ethylene-responsive transcription factor RAP2-3 (NP_188299.1)	ERF6 (NP_001266125.1)	ethylene-responsive transcription factor RAP2-3-like (NP_001287809.1)
<i>FcERF7762</i>	ethylene-responsive transcription factor 3 (NP_175479.1)	ethylene-responsive transcription factor 3-like (XP_004248374.1)	ethylene-responsive transcription factor 3-like (XP_008362296.1)
<i>FcERF4552</i>	putative AP2/EREBP transcription factor (AAT44938.1)	ERF062-like (XP_004247959.1)	ERF062 (XP_008380561.1)
<i>FcERF3418</i>	ERF003 (NP_197901.1)	ethylene response factor 1 (NP_001234848.1)	ERF003-like (XP_008378704.1)

Table S2. Primers used for transcript isolation and sequencing. All primers except *FcACSIL*-Forward* were designed according to the developing fig transcriptome. *Designed according to GenBank sequence DQ269492.1.

Gene name cDNA isolates	Forward primer (3'→5')	Reverse primer (3'→5')
<i>FcACSIL</i>	ATGGGTTTGGCGGAGAAC*	TTAAGTTCTTGCTTGGACTAAAGGTGATTG
<i>FcACS4</i>	TTCTCCGGCACCGTCTTTGACTCC	TTAACGGTCGGTTTCACGATCATCCGAT
<i>FcACO2</i>	ATGGCAATCCCAGTGATTGATTTCTCAAAGC	CTACATGGCTCTGACAGCTTGGAACCT
<i>FcACO3</i>	ATGGCTCTCCTACACCAAGCTTGC	CTAGATGACCGTTGCTCCCATTGACCAT

Table S3. Primers used for high-throughput real-time qPCR.

Gene name	Forward primer (3'→5')	Reverse primer (3'→5')
<i>FcMADS2</i>	GTATGGCGGTAGAGCATCAGATGC	GTAAGTTCCTCCAATGTTTAAGCAGGGAAG
<i>FcMADS3</i>	CTCATACTGCGGAGGCTTCTCAATC	GCATCCAGGGTGGTATGCAAGTG
<i>FcMADS4</i>	AGACTCAGGTGAACAAGGCCAGTCA	CAACTTGAGAGCGGTATCCGAGC
<i>FcMADS5</i>	GCTCTTGTCATGGACCAAGCTGG	GCGGCAGTTAATCCCATTGCTATG
<i>FcMADS6</i>	TTCACCCGCCATCCTACTCAACC	CTCCAACATTCATCCCATCTGGACC
<i>FcMADS8</i>	GTAAGTGAAGCTGCTAGGGAGCATAA	AGTGCGTTATTGCATGGTAAAGGCTC
<i>FcACSIL</i>	CGAGAGCGCTAGAAGATTGG	CTGTGCAAATCCATCCACAA
<i>FcACS2</i>	GTCGAGCTTCGGACTAGTGTC	CCTCTTCGAGCTCTCCGA
<i>FcACS3</i>	TCGTAAGCGTCATGGAGATTC	TCGTAAGCGTCATGGAGATTC
<i>FcACS4</i>	GCCTTTGCGGAAACCTTAAT	TGCTAATTGGAACACCCATCT
<i>FcACOL</i>	GAGCAGGAAACCAAAGTGGT	CCTTCATGGCTTCAAACCTAG
<i>FcACO2</i>	CAAATCCTGAAAGGCGAGAAG	GTGCCAAACACTCTTGTACCTC
<i>FcACO3</i>	GTTTGCAGTTCCTCAAAGATGA	TCCATIGCTCAGAATTTCCAC
<i>FcEOL1</i>	GTGGTATTGACGTTAGAACCCAA	TTACTTAATTGTCGAACCATACGG
<i>FcEOL2</i>	GACCTGGAGATGGTTACCCAA	TGCTTCAGTTTCCTTCTGGTT
<i>FcETR1</i>	GCAATCTGCAAGAGGTTTCGT	ATGGAGCTTCGATTCATTGG
<i>FcETR2</i>	GAATTCGTTCCAGAGGGTGA	ACCATCGCCTCGACAATAAC
<i>FcEIN4</i>	CAATGCGAATCCGGAAGTT	GAAGTTCATCCGCCAAACCT
<i>FcERS1</i>	ACCTGCCCTTTAGCCAGAAT	CAAAACCATGATCGCATAGC
<i>FcCTR1</i>	GGACCTCCCACTGATGAAGA	GACATGTCATCACCGTGACC
<i>FcCTR2</i>	CAGCTCAGGTTGTAGCAGCA	CCAGCAGATTTTGATTAAGGC
<i>FcCTR3</i>	TTGGCTACATGCTGGAATGA	CACGATGGACACAAAATCCA

<i>FcEDR1</i>	TGGCTAGGGGTATGAATTGC	TCACCAACCTTGACATTCCA
<i>FcEIN2</i>	CTATACCTGATTCACACACCGAC	CATGGGCTTTACTACGACAGG
<i>FcEBF1</i>	TTAGCCAAGCAGGGATCGT	TGAGCGTGGGTACACCAGA
<i>FcEIL1</i>	GGAGGGTCCAACCTATGGAT	GTGATGGCTGAGGCAGTGTA
<i>FcEIL2</i>	GGTTGTTATGGGAGGTAATATCC	GTCCGCAATATTGTCATTGG
<i>FcEIL3</i>	CGGTAGTGGCTCCTAAGATTA	AATCCTGATAGGGGCATTGA
<i>FcERF9168</i>	TTGTGTTGGAAGAGGCAGAG	CCGTCCAGGTACGGAATCT
<i>FcERF12185</i>	GAGTCGATCGGGAGTGAGTT	CTCGCCGTAAAAGTACTCATCA
<i>FcERF9211</i>	ACTAGTTTCGCCGCTAGCAA	ACTAGTTTCGCCGCTAGCAA
<i>FcERF9297</i>	GCAGCATTGACGATGTTTCCAG	GCAAGGAATGTTGAGAGCTT
<i>FcERF8231</i>	CTTTTTGAAGGTGGCGAACC	TTGATTCTGGGAGGCTCTTC
<i>FcERF5086</i>	CACCGTCCTACGTTTCCAAT	AACCTCCCAAATCAAATCAGG
<i>FcERF9572</i>	GAAGAAGAATTACAGAGGAGTGAGG	GCCGTGTTAAAAGTCCCG
<i>FcERF9690</i>	ACGGAGAGAGCACGATGG	CTGAACCGGAATTCGTGACT
<i>FcERF9109</i>	GCCGGAGGAGAGTTTATTGA	ACGACGGCGTATTGATCCT
<i>FcERF11513</i>	TGGGAGGAGGGTGTTC	CTTGTTTCGGCTCACGGAG
<i>FcERF11632</i>	GGGAGGAGGGTATTCAAGGA	GACTTGGTATTTCGGCTCTCG
<i>FcERF12049</i>	GTGGGAGAGTTTTGATTTTTCC	TTCTCGGTTTCGTCTCGGA
<i>FcERF8090</i>	GAGTCAGACATCTCTTGCCG	AACCAGACCCGCTTTCTC
<i>FcERF10238</i>	GTTAGATTCCCGTTCCAGCA	GTTGATACGGGACGCTCTTG
<i>FcERF10715</i>	GCTTAGCAGACCGGATTCA	TTCAACCACGGAAGACGAGT
<i>FcERF998</i>	ATGCACCACTGGATGCCTT	AGGAAAGCAGGGGAAGTAGC
<i>FcERF14722</i>	GAATTCTCGTTGTAGCTTTAGGG	GTCGGGAAGGTGCCTAGC
<i>FcERF2127</i>	ATATGGTCATGTCAACCGCA	GGTTTTGAATTAGCACCAACG
<i>FcERF9816</i>	CGAACAAGAGGTCGAGGATT	AAGATGAGGTCGGGGAAGTT

<i>FcERF14757</i>	CAGCCCATCAACACGACC	CTAGCCAGAGGCGAGTCCT
<i>FcERF8610</i>	ACGATGGTGGAAGTGAGAGG	TTGGTTAGCAATGGTGCAGAC
<i>FcERF3350</i>	CGCATCCGAACCCAATTC	AATCTTCCGTGGCTCAGATC
<i>FcERF9006</i>	CGGACCTCTACCACTATGCAC	TGTAATGAAAGGGAGCTGCTG
<i>FcERF1362</i>	TGTCGCCACCTAACAAAAGTG	TGTTCTTGCGAGTTCGTTGA
<i>FcERF7762</i>	CGGTCCAAGAAGTAAGATTCAGA	GAAGGTGCCGAGCCAGA
<i>FcERF4552</i>	AAGGCGTCTCGGGTTCTC	AAACCATTGGCTTTCGAGTC
<i>FcERF3418</i>	CGAAACCCGTCGAAACG	GGTTGCTCTCAACTTAAACCTTC

Fig. S1. Newly isolated *FcACSIL* transcript alignment with published sequence of *FcACSI*, DQ269492.1 (Owino *et al.*, 2006). The frame-shift position is marked by a red asterisk. Alignment was performed by MUSCLE program using default parameters (<http://www.ebi.ac.uk/Tools/msa/muscle>) (Edgar, 2004).

FcACS1 DQ269492.1	1	ATGGGTTTGGCGGAGAACCAGCTCTGCTTTGATTTGGTTCAAGACTGGATTCTGAACAC	60
FcACS1L	1	ATGGGTTTGGCGGAGAACCAGCTCTGCTTTGATTTGGTTCAAGACTGGATTCTGAACAC	60
FcACS1 DQ269492.1	61	CCCAGAGCCTCCATTTGCACGCCTGAAGGAGTTGATGAATTCAGAGACATAGCTATTTTC	120
FcACS1L	61	CCCAGAGCCTCCATTTGCACGCCTGAAGGAGTTGATGAATTCAGAGACATAGCTATTTTC	120
FcACS1 DQ269492.1	121	CAAGACTACCATGGCCTGCCAGAGTTCGAAAAAGCTGIAGCGGAGTTCATGGGACAGGTT	180
FcACS1L	121	CAAGACTACCATGGCCTGCCAGAGTTCGAAAAAGCTGIAGCGGAGTTCATGGGACAGGTT	180
FcACS1 DQ269492.1	181	AGAGGAAACCGAGTAACCTTTCCATCCCGACCGCATTGIAATGAGCGGAGGAGCGACCGGT	240
FcACS1L	181	AGAGGAAACCGAGTAACCTTTCCATCCCGACCGCATTGIAATGAGCGGAGGAGCGACCGGT	240
FcACS1 DQ269492.1	241	GCTCATGAGATGGTTACTTTTTGCTTGGCTGATCCAGGAGAAGCATTCTGGTTCCCTGTT	300
FcACS1L	241	GCTCATGAGATGGTTACTTTTTGCTTGGCTGATCCAGGAGAAGCATTCTGGTTCCCTGTT	300
FcACS1 DQ269492.1	301	CCTIATTATCCAGGATTCACCGAGACTTGAGATGGAGAACGGGAGTACAACIAATTCCG	360
FcACS1L	301	CCTIATTATCCAGGATTCACCGAGACTTGAGATGGAGAACGGGAGTACAACIAATTCCG	360
FcACS1 DQ269492.1	361	GTCCACAGCGAAAGCGCAACAACTTCAAGGTGACAAGGGAAGCCTTGGAAAGAACCCAT	420
FcACS1L	361	GTCCACAGCGAAAGCGCAACAACTTCAAGGTGACAAGGGAAGCCTTGGAAAGAACCCAT	420
FcACS1 DQ269492.1	421	GAGAAGGCTCAGAGGAGAACATAAGAGTGAAGGTTTGGCTGATCACCAATCCATCAAAC	480
FcACS1L	421	GAGAAGGCTCAGAGGAGAACATAAGAGTGAAGGTTTGGCTGATCACCAATCCATCAAAC	480
FcACS1 DQ269492.1	481	CCCTTGGGCACCATCCTIAGACAGAGACACIAAGGATGATAGTGGGTTTCATCAACGAG	540
FcACS1L	481	CCCTTGGGCACCATCCTIAGACAGAGACACIAAGGATGATAGTGGGTTTCATCAACGAG	540
FcACS1 DQ269492.1	541	AAGAGCATCCACCTAGTCTGCGACGAAATCIATGCTGCCACGGTCTTCAACCAGCCGCCA	600
FcACS1L	541	AAGAGCATCCACCTAGTCTGCGACGAAATCIATGCTGCCACGGTCTTCAACCAGCCGCCA	600
FcACS1 DQ269492.1	601	TTCGTGAGCATCTCGGAGATCAIAGCAGAGGATACAGACATCGAGTGCACACAGACCTC	660
FcACS1L	601	TTCGTGAGCATCTCGGAGATCAIAGCAGAGGATACAGACATCGAGTGCACACAGACCTC	660
FcACS1 DQ269492.1	661	ATCCACATTGTTIACAGCCTCTCAAGGACATGGGATTCCCTGGCTTCAGAGTCGGCGTT	720
FcACS1L	661	ATCCACATTGTTIACAGCCTCTCAAGGACATGGGATTCCCTGGCTTCAGAGTCGGCGTT	720
FcACS1 DQ269492.1	721	GTCTACTCCTIACAACGACGCGGTGGTGAATTGCGCGCGAAAGATGTCAAGCTTCGGATIA	780
FcACS1L	721	GTCTACTCCTIACAACGACGCGGTGGTGAATTGCGCGCGAAAGATGTCAAGCTTCGGATIA	780
FcACS1 DQ269492.1	781	GTCTCGTCTCAAACACAACACCTGATTGCAGCCATGTTGTGGATGATGTGTTTGTGACG	840
FcACS1L	781	GTCTCGTCTCAAACACAACACCTGATTGCAGCCATGTTGTGGATGATGTGTTTGTGACG	840
FcACS1 DQ269492.1	841	AGGTTTTTGGCCGAGAGCGCTAGAAGATTGGAAGAAGGCACAACGGCTTACCCGAGGA	900
FcACS1L	841	AGGTTTTTGGCCGAGAGCGCTAGAAGATTGGAAGAAGGCACAACGGCTTACCCGAGGA	900
FcACS1 DQ269492.1	901	CTCGCTCAGGTTIAGAATTIACCTGCTTGGAGAGCAATGCAGGCCTGTTTTTGTGGATGGAT	960
FcACS1L	901	CTCGCTCAGGTTIAGAATTIACCTGCTTGGAGAGCAATGCAGGCCTGTTTTTGTGGATGGAT	960
FcACS1 DQ269492.1	961	TTGCACAGGCTTCTCAAAGAAACAGACACTTGATGCAGAGATGGCTCTATGGAAAATCATC	1020
FcACS1L	961	TTGCACAGGCTTCTCAAAGAAACAGACACTTGATGCAGAGATGGCTCTATGGAAAATCATC	1020
FcACS1 DQ269492.1	1021	ATTAAATGAAGTIAAGCTCAACGTTTCGCCCGGTTTTTCGTTTCATTGCCCCGAACCGGG	1079
FcACS1L	1021	ATTAAATGAAGTIAAGCTCAACGTTTCGCCCGGTTTTTCGTTTCATTGCCCCGAACCGGG	1080
FcACS1 DQ269492.1	1080	TGGTTCAGCGTATG-----	1093
FcACS1L	1081	TGGTTCAGCGTATG-----	1140
FcACS1 DQ269492.1	1093	-----	1093
FcACS1L	1141	AIACGAAACTTTGTGCTCCAAAAGAGGAGCTTGTGTGAAGCCAAGTAAAGAAAGTGC	1200
FcACS1 DQ269492.1	1093	-----	1093
FcACS1L	1201	TGGCAAGATCACAACTCAGGCTCAGCTTGAAGCTCGCCGATTTCAGCATTCTTGATG	1260
FcACS1 DQ269492.1	1093	-----GTTCCCTGCAGTTA-----	1109
FcACS1L	1261	TCGCCTCATTCTCCIATTTCTCAATCACTTTAGTCCAAAGCAAGAACTTAA	1311

Fig. S2. Phylogenetic analysis of *FcACS* predicted proteins. The three types of ACS proteins are noted. Amino acid sequences of ACSs from tomato and apple were obtained from the NCBI database (<http://www.ncbi.nlm.nih.gov/>). NCBI accession numbers of the proteins are as follows: SIACS7 (AAK72432.1), SIACS8 (NP_001234160.1), SIACS3 (NP_001234026.1), MdACS1 (ABA03056.1), SIACS5 (NP_001234156.1), MdACS3a (AAB67989.1), MdACS3b (BAE94691.1), MdACS3c (BAE94692.1), SIACS2 (NP_001234178.1), SIACS4 (NP_001234280.1), MdACS5a (NP_001280768.1), MdACS5b (NP_001280870.1), SIACS1a (AAF97614.1), SIACS1b (AAF97615.1), SIACS6 (AAK72433.1) and PgACS1 (ABM60747.1)*. Alignment was performed by MUSCLE program using default parameters (<http://www.ebi.ac.uk/Tools/msa/muscle>) (Edgar, 2004). A phylogenetic tree was reconstructed based on the maximum likelihood (ML) framework using PhyML software (Guindon and Gascuel, 2003) based on the JTT matrix-based model (Jones *et al.*, 1992). A bootstrap consensus tree was inferred from 1,000 replicates and the number of replicate trees in which the associated clade clustered together in the bootstrap test is designated next to the branches (Felsenstein, 1985). The tree was graphically designed with the use of Fig Tree version 1.4 (<http://tree.bio.ed.ac.uk/software/figtree/>). *PgACS1 (*Picea glauca* L. ACS1) was defined as an outgroup.

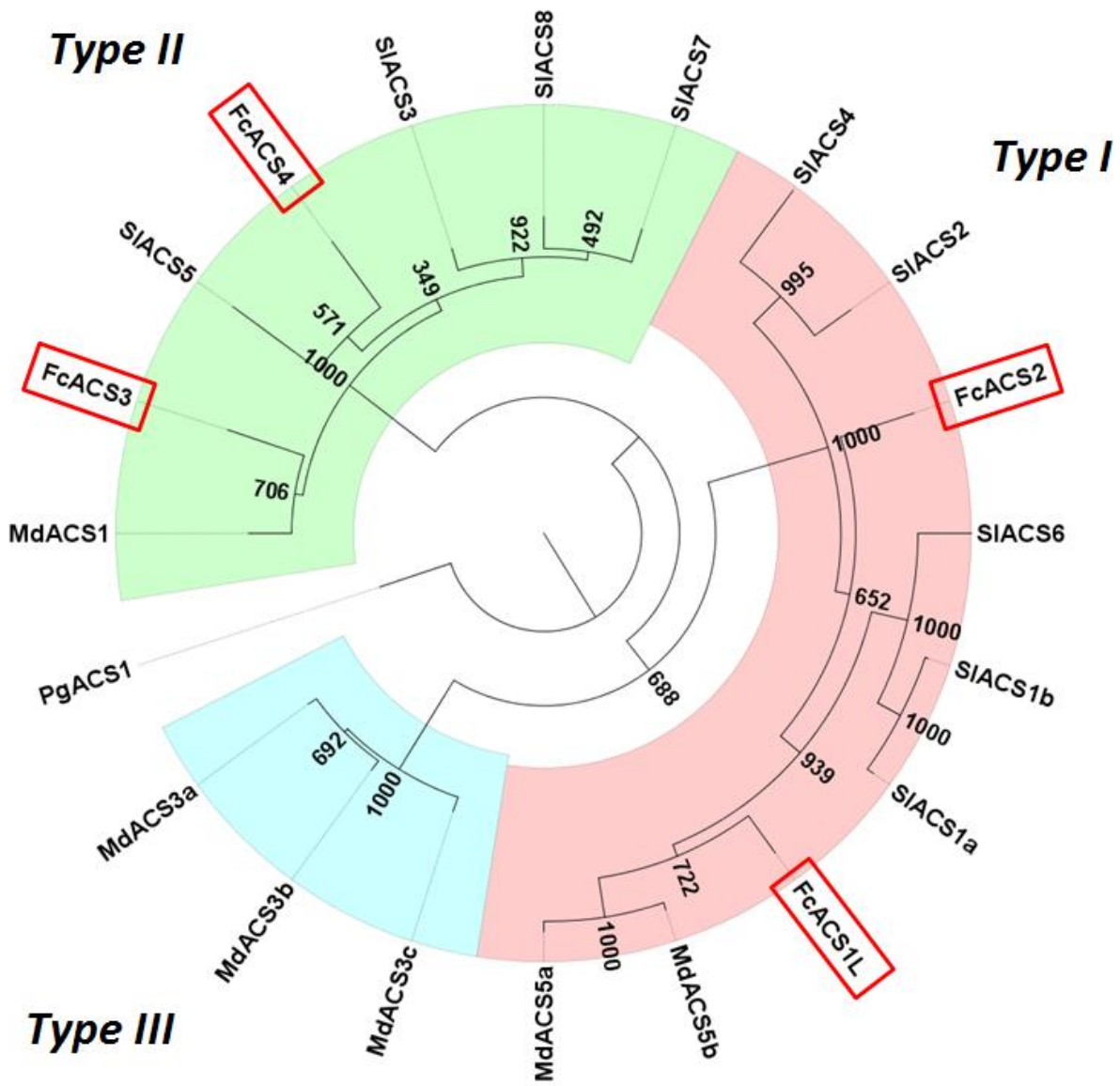


Fig. S3. Isolated *FcACOL* transcript (published previously under NCBI accession number AB307720.1 2007) aligned with the published sequence of *FcACO1* (Owino *et al.*, 2006). Alignment was performed by MUSCLE program using default parameters (<http://www.ebi.ac.uk/Tools/msa/muscle>) (Edgar, 2004).

```

FcACOL          1  ATGGAGCAGATCACTTTTCCAGTTATTGACATGACAAAACCTCAACGGCGACCAGAGAAAAG 60
FcACO1|DQ269495.1 1  ----CGCCGATC----- 8

FcACOL          61  ACAACCATGGAATTGATAAACGAAAGCTTGTGAGAAATGGGGCTTCTTTGAGGTGATGAA 119
FcACO1|DQ269495.1 8  -----CGCTTGTGAGAAATGGGGCTTCTTTGAGGTGATGAA 45

FcACOL          120  CCATGGGATATCCCACGAGCTCATGGACACGGTGGACAGGCTGACAAAGGAGCACTACAG 179
FcACO1|DQ269495.1 46  CCATGGGATATCCCACGAGCTCATGGACACGGTGGACAGGCTGACAAAGGAGCACTACAG 105

FcACOL          180  AAAGTGCATGGAACAAAGGTTCAAGGAAATGGTGGCAACCAAGGGACTTGATGTTGTTCA 239
FcACO1|DQ269495.1 106  AAAGTGCATGGAACAAAGGTTCAAGGAAATGGTGGCAACCAAGGGACTTGATGTTGTTCA 165

FcACOL          240  GTCTGAAATCAAGGACTTGGACTGGGAAAGCACCTTCTTCTTGCGCCACCTTCCCCTCTC 299
FcACO1|DQ269495.1 166  GTCTGAAATCAAGGACTTGGACTGGGAAAGCACCTTCTTCTTGCGCCACCTTCCCCTCTC 225

FcACOL          300  AAACATGTCCGAAATCCCCGATCTCGGCGACGACTACAGGAAGGTAATGAAGCAATTTGC 359
FcACO1|DQ269495.1 226  AAACATGTCCGAAATCCCCGATCTCGGCGACGACTACAGGAAGGTAATGAAGCAATTTGC 285

FcACOL          360  TCGGCACTGCGACAACTGGCAGAGCATCTCTTGGACTTGCTGTGTGAGAAATCTTGGACT 419
FcACO1|DQ269495.1 286  TCGGCACTGCGACAACTGGCAGAGCATCTCTTGGACTTGCTGTGTGAGAAATCTTGGACT 345

FcACOL          420  AGACAAGGGTIACATAAAGAGCGTGTTCIATGGGTCAAAGGGTCCAAATTTTGGAAACCAA 479
FcACO1|DQ269495.1 346  AGACAAGGGTIACATAAAGAGCGTGTTCIATGGGTCAAAGGGTCCAAATTTTGGAAACCAA 405

FcACOL          480  GGTGAGTAACTACCTCCATGCCCAAACCAGACCTGATCAAGGGCCTCCGGGCCACAC 539
FcACO1|DQ269495.1 406  GGTGAGTAACTACCTCCATGCCCAAACCAGACCTGATCAAGGGCCTCCGGGCCACAC 465

FcACOL          540  CGATGCCGGCGGCATCACTTGTCTTCCAGACGACAAGGTCAACGGTCTTCAGCTCCT 599
FcACO1|DQ269495.1 466  CGATGCCGGCGGCATCACTTGTCTTCCAGACGACAAGGTCAACGGTCTTCAGCTCCT 525

FcACOL          600  CAAGGATGACCAGTGGATTGATGTCCCTCCAATGCGCCACTCCATCGTCATCAACTTGGG 659
FcACO1|DQ269495.1 526  CAAGGATGACCAGTGGATTGATGTCCCTCCAATGCGCCACTCCATCGTCATCAACTTGGG 585

FcACOL          660  TGAACAACCTTGAGGTAATCACCAAATGGGGCAIACAAGAGCGTGATGCACCGTGTGGT 718
FcACO1|DQ269495.1 586  TGAACAACCTTGAGGTAATCACCAAATGGGGCAIACAAGAGCGTGATGCACCGTGTGGT 644

FcACOL          719  CTCAACCGGACGGTAAACAGGATCTCCCIAGCATCGTCTIACACCCGGGAGCGACGG 778
FcACO1|DQ269495.1 645  CTCAACCGGACGGTAAACAGGATCTCCCIAGCATCGTCTIACACCCGGGAGCGACGG 704

FcACOL          779  TGATCTACCCAGCTCCGGCAATTGGTGAATAAGCGCGGCAACAGAAAACCAAAGTGGTG 837
FcACO1|DQ269495.1 705  TGATCTACCCAGCTCCGGCAATTGGTGAATAAGCGCGGCAACAGAAAACCAAAGTGGTG 764

FcACOL          838  TACCCAAAGTTTCGTGTTTATGACTACATGASGCTGIATGCCCGGTTGAAATTCAGGCA 897
FcACO1|DQ269495.1 765  TACCCAAAGTTTCGTGTTTATGACTACATGASGCTGIATGCCCGGTTGAAATTCAGGCA 823

FcACOL          898  AAGAGCCCTAGGTTTGAAGCCATGAAGGCCGTGGAAACCCCACTAAACGTGGATCCATT 957
FcACO1|DQ269495.1 824  AAGAGCC-----CCTTCAATTAATCAATTC----- 850

FcACOL          958  GCAACTGTTTGA 969
FcACO1|DQ269495.1 850  ----- 850

```

Fig. S4. Alignment of deduced FcMADS8 amino acid sequence and SIRIN protein (NCBI accession number ABJ98752.1). Marked above sequence: red line – MADS-box domain; blue line – K-box domain; green line – C terminus. Black highlighted amino acids – identical residues; gray highlighted – similar residues. Alignment was performed by MUSCLE program using default parameters (<http://www.ebi.ac.uk/Tools/msa/muscle>) (Edgar, 2004).

```

SIRIN      1  MGRGFVELRRIENRINRÇVTFARRRNGLLRFAYELSLCTAFETALIFSSRGRLYEFCNSNSMSTLEERYHYNYGTLEG 80
FcMADS8   1  MGRGFVELRRIENRINRÇVTFARRRNGLLRFAYELSVLCDSFVGLIIFSSRGRLYEFCSGHSMARTTEREFESYNYGQ-EG 79

SIRIN     81  TOTSSDSQNNYQEYLRLRTRFVELLQSSQRHLLGEDLQQLGTRDLEQLERQLDSSLRQIRSTKIQHILDQLAELQQKECSL 160
FcMADS8   80  KQPEKDSQSSYQDYLRLRKRVELLRRTRQRHLLGEDLEQLGLRRLGQLEHRLDVSLRQIRSTKIQSMLDQLSNLRKREESL 159

SIRIN    161  TEANKSLRHRLEELCVTFQTSWHCGEQSVQYRHEQESHHEGF-FOEVMNCNNQLETSIFYDINVQPENAAPSTHDATGVVVG 239
FcMADS8  160  LEANNEIKRRLEEMGVSEAAAR---EHNIAVNAHHLQPMDESEEPLECNNALEISVNSAVRGEVNNV-----G 226

SIRIN    240  WML 242
FcMADS8  227  FDL 229

```

References

- Edgar RC.** 2004. MUSCLE: multiple sequence alignment with high accuracy and high throughput. *Nucleic Acids Research* **32**, 1792–1797.
- Felsenstein J.** 1985. Confidence limits on phylogenies: an approach using the bootstrap. *Evolution* **39**, 783–791.
- Guindon S, Gascuel O.** 2003. A simple, fast, and accurate algorithm to estimate large phylogenies by maximum likelihood. *Systematic Biology* **52**, 696–704.
- Jones DT, Taylor WR, Thornton JM.** 1992. The rapid generation of mutation data matrices from protein sequences. *Computer Application in the Biosciences* **8**, 275–282.
- Owino WO, Manabe Y, Mathooko FM, Kubo Y, Inaba A.** 2006. Regulatory mechanisms of ethylene biosynthesis in response to various stimuli during maturation and ripening in fig fruit (*Ficus carica* L.). *Plant Physiology and Biochemistry* **44**, 335–342.