

Figure S1. Differential expression analysis and enrichment analysis among sequencing samples. **(A)** Volcano map of DGEs between T02 and T01. **(B)** Volcano map of DGEs between T03 and T01. **(C)** Volcano map of DGEs between T03 and T02. **(D)** Venn diagram of co-DGEs among the samples. **(E)** GO classification and the enrichment analysis of DEGs. **(F)** KEGG enrichment analysis of DEGs. T01, diseased peels of calcium-deficient apples; T02, healthy peels of calcium-deficient apples; T03, peels of calcium-sufficient apples. Negative \log_{10} P values indicate the y-axis, and \log_2 -fold changes indicate the x-axis.

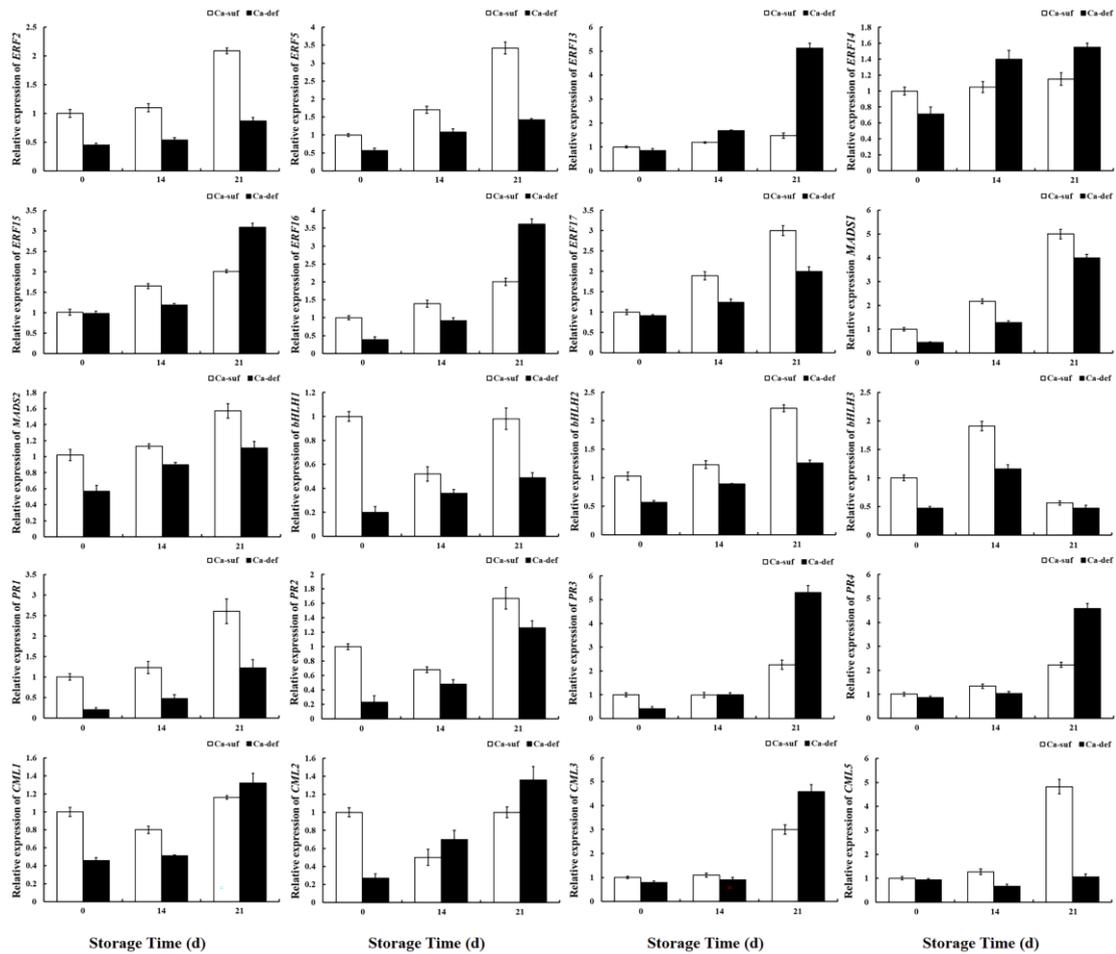


Figure S2. Expression pattern analysis of DEGs (*ERF2*, *ERF5*, *ERF13*, *ERF14*, *ERF15*, *ERF16*, *ERF17*, *MADS1*, *MADS2*, *bHLH1*, *bHLH2*, *bHLH3*, *PR1*, *PR2*, *PR3*, *PR4*, *CML1*, *CML2*, *CML3* and *CML5*) in apple peels under calcium sufficiency and deficiency with storage at 0, 14, 21 DAS. Ca-suf, Ca sufficiency; Ca-def, Ca deficiency. Data are presented as means \pm SD (n = 3). Gene expression of *MdUBQ* (*UBQ*, accession number MDU74358) was used as a normalization gene, and relative gene expression was determined using the $2^{-\Delta\Delta CT}$ method to calculate.

Figure S3

Gene name	LOG2 fold change			RT-qPCR		
	Ca-suf	Ca-def	Ca-suf/Ca-def	Ca-suf	Ca-def	Ca-suf/Ca-def
ERF2	628.97	143.02	4.40	4.56	2.27	2.01
ERF5	121.28	9.72	12.48	4.05	2.66	1.52
ERF13	0.54	60.54	0.01	1.88	2.87	0.66
ERF14	6.77	75.66	0.09	3.27	3.81	0.86
ERF15	1.66	71.69	0.02	2.48	4.32	0.57
ERF16	4.24	398.90	0.01	0.96	1.24	0.77
ERF17	177.42	12.83	13.83	4.24	2.46	1.72
MADS1	270.98	86.53	3.13	10.39	9.08	1.14
MADS2	228.16	59.12	3.86	10.85	6.58	1.65
bHLH1	249.49	25.61	9.74	0.62	0.29	2.14
bHLH2	658.29	25.99	25.33	7.11	4.53	1.57
bHLH3	781.14	37.29	20.95	0.22	0.11	2.00
PR1	197.82	88.19	2.24	2.70	1.32	2.05
PR2	21.90	102.37	0.21	1.75	1.34	1.31
PR3	10.39	401.69	0.03	2.66	5.79	0.46
PR4	17.37	3935.32	0.00	8.82	13.36	0.66
CML1	6.33	108.69	0.06	15.03	20.82	0.72
CML2	15.58	201.79	0.08	1.01	1.38	0.73
CML3	10.74	252.54	0.04	5.46	7.82	0.70
CML5	108.54	27.91	3.89	8.16	6.92	1.18

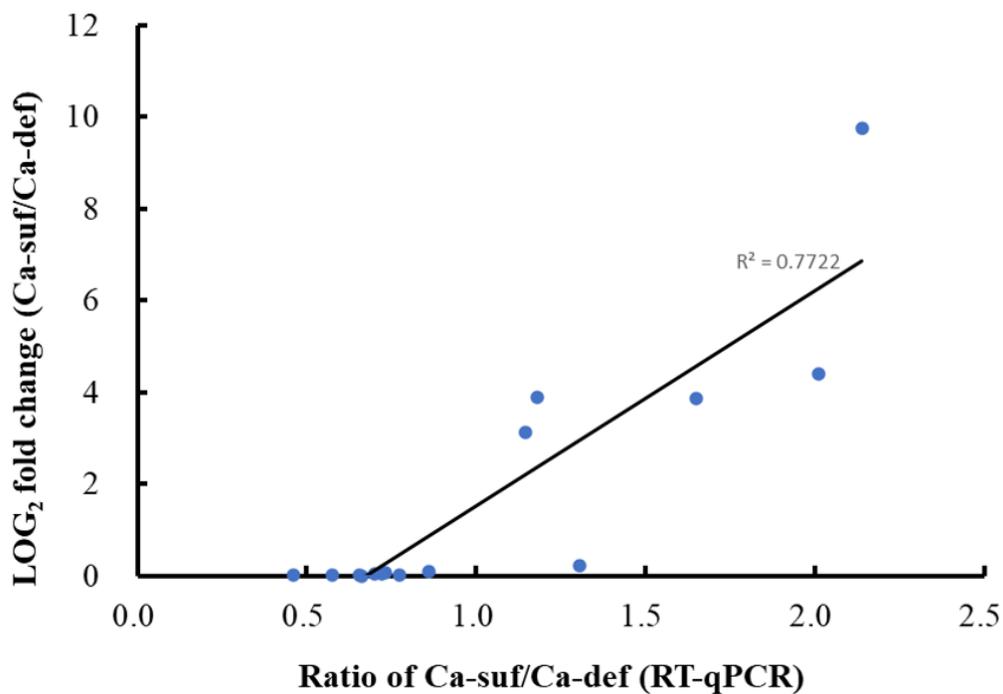


Figure S3. Correlation analysis of the RNA-seq results and RT-qPCR results for the DEGs.

Table S1. Real-time qPCR primers used in this study.

Gene name	Gene ID	Primer sequence (5'-3')
<i>MdERF2</i>	MD13G1163300	F-GTTGTTGTTTCGCTGGTGATG R- CCTTCCCTCATAATGACGATCC
<i>MdERF5</i>	MD17G1226700	F- GCCGGTTCCTTGCTTCATTTT R- CCCACAGATGAGCTTCGTATC
<i>MdERF13</i>	MD14G1147100	F-GGTTGAATCGGAGGAGAAAGAG R- CACCTGAGCTAGAGCTGAAAC
<i>MdERF14</i>	MD16G1140800	F-GAAGCATCAGAGGAGAAAGAGAG R- CTTGAGTGAGAGCAGAAACCA
<i>MdERF15</i>	MD07G1248400	F-CAGCTACTGCATCTCCTACTTG

<i>MdERF16</i>	MD06G1130400	R- GTCCTCCCACCTGATACTGATA F- CCGAAAGAGTTCAGGGAAGAAT
<i>MdERF17</i>	MD01G1177200	R- GTGGTGGTCCGTAATACTTTGA F- CGAGTACTCATCCCTCTCTGAA
<i>MdbHLH1</i>	MD13G1150900	R- CGGGTTTTCGTTTTCGGAATTTG F- TCTGATTTTGAATCCCGTAGC
<i>MdbHLH2</i>	MD06G1133200	R- GGTGGAGGTGGTCTTGTTT F- CTCTCGGTGAAACTTGACTCTT
<i>MdbHLH3</i>	MD17G1049300	R- GTGACGTGGGACTGAACATAA F- CATGTCTAGCAGAGGGTCAAG
<i>MdMADS1</i>	MD06G1204300	R- CTCAGGAACCAGTTGACGTAAT F- AGAACCCCTGCATGTGCTTATAG
<i>MdMADS2</i>	MD16G1058500	R- TGAGACAACCTACGCTGAAAC F- GGCACGGACCAAGTACATAATA
<i>MdPR1</i>	MD10G1017300	R- GAGATTTACTACACCAGGCTTCTC F- ACCGTTCTATATGACTGGAGA
<i>MdPR2</i>	MD10G1286300	R- AGAGAGAGAGAGAGAGACCTTTG F- CACATCCATCGTCAACCAAATC
<i>MdPR3</i>	MD05G1017400	R- TGCAGAGTCACGGATTTTCAG F- ATGGCCTCACCAAGAGTTAAG
<i>MdPR4</i>	MD05G1110000	R- GTTGACAGCACCATAGTTAGGA F- GACTCATGCCCAAGACACA
<i>MdCML1</i>	MD12G1087300	R- CTATATTGTCCTCCCACGTCAAG F- CGTCATCCTCTTCCTCATAAC
<i>MdCML2</i>	MD06G1231000	R- GACCACTCACCCGATCTTTC F- CAAAGACGCCTTCGATCTCTAC
<i>MdCML3</i>	MD04G1178900	R- ATCGTGCAGGCCATGTT F- CGCCGCATCTTTGATATGTTT
<i>MdCML4</i>	MD16G1169000	R- CAGTGTCTAACCCGAGAAGTG F- GTTCCTCTGGTGGATGGATTC
<i>MdCML5</i>	MD08G1043100	R- CTCGTTCCATCCGTGCATAA F- ACGACGGATCGATTACAAG
<i>MdDCD1</i>	MD14G1242800	R- AGCACGACAACCTCCAATCTC F- CTGTGACGACCCTGATTACTTC
<i>MdDCD2</i>	MD06G1235700	R- CCCAGACCTTTGGCATTCT F- ACACCAAGAACAGTACGATAAC
<i>MdDCD3</i>	MD06G1235800	R- GGTCATCACGTTGTCCAAGA F- ATCTCCTGGTTGAGCGTTTAG
<i>MdDCD4</i>	MD13G1105700	R- CTCAGAGGTGGTAACTGCATAG F- CTCCAACATTACCAACCCATCT
<i>MdLCD1</i>	MD12G1200100	R- TCCAACCTTTTCGCGCCTTATTA F- ACCGCTCTTCTCTCTCTCTT
<i>MdLCD2</i>	MD04G1186700	R- CCATTTGGCGATTAGGGTTTG F- GTTGCACCATCCTGTTGTTTC
<i>MdLCD3</i>	MD15G1421300	R- TTGCGGGCTATAATCCCTATTC F- CATCTCGCCGTCTACTTCTTG
<i>MdSiR1</i>	MD05G1081500	R- GTCCTTACGCTTCTTTCAGTT F- TCCTCGTCTCTCTCTCAATTCC
<i>MdOASTL1</i>	MD06G1007300	R- GGGATGGTGACATGGTTGATAG F- AGTTTCGACAACCCCTGCTAATC
<i>MdTUB</i>	GO562615	R- AACCGTCCTACACCAGAAATG F- ACACGGGGAGGTAGTGACAA
<i>MdUBQ</i>	MDU74358	R- CCTCCAATGGATCCTCGTTA F- CTCGTTGGTGGTTTTTAAAGT
		R- GGAGGCAGAAACAGTACCAT

Table S2. Primers used in this study for developing the constructs.

The purpose	The primers	The sequences of the primer (5'-3')
For construction of re-combinant vector of pSAK277	<i>ERF2-EcoRI-F</i>	actagtggatccaagaattcATGTGTGGTGGTGCTATCATTTC
	<i>ERF2-XbaI-R</i>	tcattaaagcaggactctagaTTAATACAGAAGCTGCCCTTGTTG
	<i>ERF17-EcoRI-F</i>	actagtggatccaagaattcATGGTAATTAGCTTCCCCTTCTGC
	<i>ERF17-XbaI-R</i>	tcattaaagcaggactctagaTCAAGCACCTTCTCCTTCCCA
	<i>bHLH2-EcoRI-F</i>	actagtggatccaagaattcATGGCAGCCTTTTCATACCAA
	<i>bHLH2-XbaI-R</i>	tcattaaagcaggactctagaTTAATTGAAAGAACACAAGTTGCTGT
For construction of pGreen II 0800-LUC binary vector	<i>CML5-HindIII-F</i>	gtcgacggtatcgataagcttTAAAGGTATGAGCACGGAAACTGA
	<i>CML5-BamHI -R</i>	cgctctagaactagtgatccTGCTGTCGTCTGCAACAATTCA