

Supplementary Informations

Soybean Calmodulin-binding Transcription Activators, GmCAMTA2 and GmCAMTA8, Coordinate the Circadian Regulation of Developmental Processes and Drought Stress Responses

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Supplementary Materials:

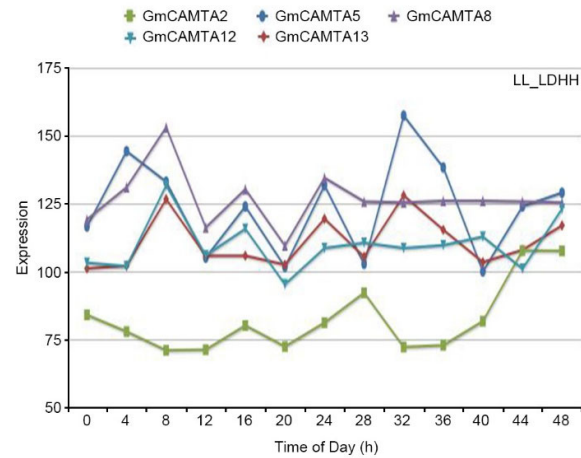
Supplementary Figure S1. Diurnal expression of *GmCAMTAs* in the Diurnal web database.

Supplementary Figure S2. Transcriptional expression of *GmCAMTAs* in soybean tissues using qRT-PCR analysis.

Supplementary Figure S3. RT-PCR analysis in *Arabidopsis* transgenic plants overexpressing *GmCAMTA2* and *GmCAMTA8*.

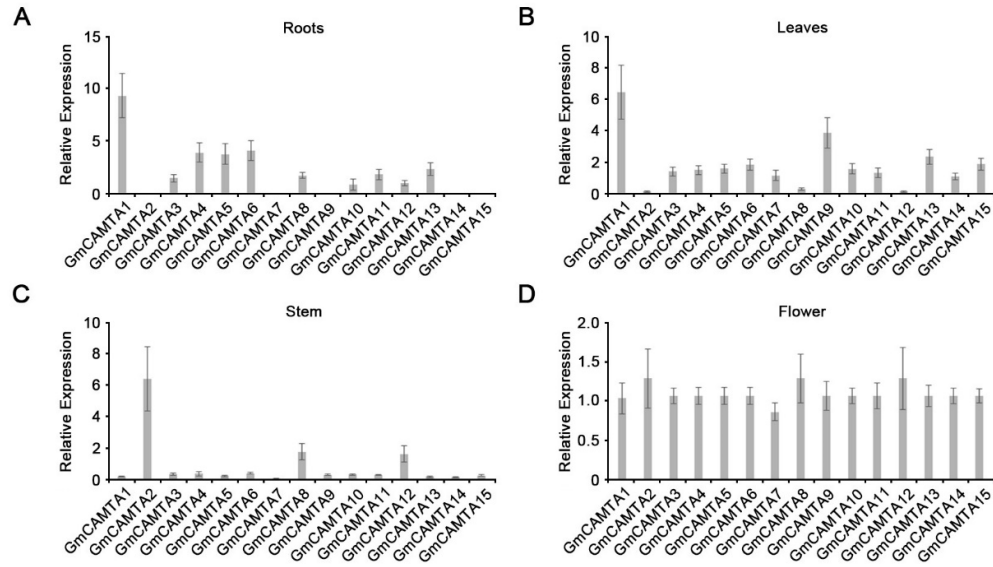
Supplementary Table S1. List of 15 *GmCAMTA* genes in the Phytozome database.

Supplementary Table S2. List of primers in this study.

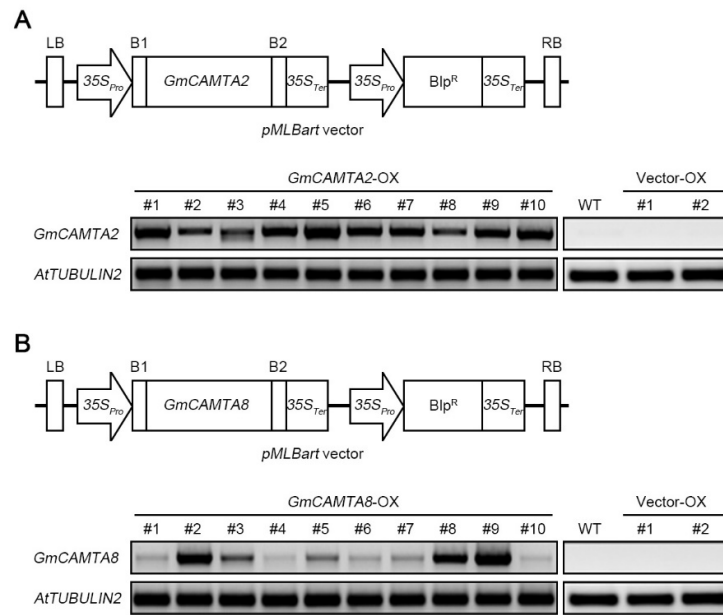


Supplementary Figure S1. Diurnal expression of *GmCAMTAs* in the Diurnal web database.

The diurnal expression of *GmCAMTA2*, *GmCAMTA5*, *GmCAMTA8*, *GmCAMTA12*, and *GmCAMTA13* under LL_LDHH condition using the Diurnal database tool (<http://diurnal.mocklerlab.org/>). The diurnal expression data of other *GmCAMTAs* were not available in the Diurnal web database. The “LL_LDHH” means a constant light for 24 h (LL), light 12h/dark 12h conditions (LD), and a constant hot temperature (31 °C; HH).



Supplementary Figure S2. Transcriptional expression of GmCAMTAs in soybean tissues using qRT-PCR analysis. The expression levels of *GmCAMTAs* were analyzed in different tissues of soybean plants using qRT-PCR. The total RNA was extracted from various tissues, including (A) roots, (B) leaves, (C) stem, and (D) flower, at the V4 stages of WT soybeans grown under long-day conditions (16h light / 8h dark). qRT-PCR analysis was performed with each *GmCAMTA*-specific primer (Supplementary Table S2). *GmTUBULIN* was used as an internal control for normalization. Error bars represent the SD of three biological replicates, each with three technical replicates.



Supplementary Figure S3. RT-PCR analysis in *Arabidopsis* transgenic plants overexpressing *GmCAMTA2* and *GmCAMTA8*. Upper panels in (A) and (B); Schematic representation of the *pMLBart* binary vectors used for generating *Arabidopsis* transgenic plants overexpressing *GmCAMTA2* (A) and *GmCAMTA8* (B). Bottom panels in (A) and (B); Analysis of RT-PCR. Total RNAs isolated from each 10-day-old seedling of *Arabidopsis* T1 transgenic plants overexpressing *GmCAMTA2* (A) and *GmCAMTA8* (B). The analysis of RT-PCR was performed with each *GmCAMTA*s-specific primer (Supplementary Table S2). The *AtTUBULIN2* in *Arabidopsis* was used as an internal control.

Supplementary Table S1. List of 15 *GmCAMTA* genes in the Phytozome database.

Gene Name	Locus No.	Chromosome No.	Genome Length (bp)	Transcript Length (bp)	CDS Length (bp)	Protein Length (aa)
<i>GmCAMTA1</i>	<i>Glyma.05G178200</i>	5	8,069	3,889	3,369	1,123
<i>GmCAMTA2</i>	<i>Glyma.08G135200</i>	8	8,007	3,872	3,309	1,103
<i>GmCAMTA3</i>	<i>Glyma.15G053600</i>	15	11,253	3,830	3,267	1,089
<i>GmCAMTA4</i>	<i>Glyma.08G072100</i>	8	11,294	3,813	3,240	1,080
<i>GmCAMTA5</i>	<i>Glyma.05G117000</i>	5	10,456	3,791	3,267	1,089
<i>GmCAMTA6</i>	<i>Glyma.08G178900</i>	8	11,604	3,947	3,246	1,082
<i>GmCAMTA7</i>	<i>Glyma.17G038800</i>	17	8,414	3,665	3,000	1,000
<i>GmCAMTA8</i>	<i>Glyma.15G143400</i>	15	9,976	3,274	2,736	912
<i>GmCAMTA9</i>	<i>Glyma.09G038300</i>	9	10,333	3,378	2,736	912
<i>GmCAMTA10</i>	<i>Glyma.05G148300</i>	5	10,592	3,637	2,952	984
<i>GmCAMTA11</i>	<i>Glyma.18G005100</i>	18	9,037	3,570	2,889	963
<i>GmCAMTA12</i>	<i>Glyma.17G031900</i>	17	8,109	3,606	2,769	923
<i>GmCAMTA13</i>	<i>Glyma.07G242000</i>	7	8,858	3,425	2,766	922
<i>GmCAMTA14</i>	<i>Glyma.11G251900</i>	11	8,300	3,196	2,733	911
<i>GmCAMTA15</i>	<i>Glyma.08G105200</i>	8	8,020	3,220	2,898	966

Supplementary Table S2. List of primers in this study.

Gene	Primer	Direction	Sequence (5'→3')	Purpose
<i>GmCAMTA1</i> (<i>Glyma.05G178200</i>)	MG-2943	Forward	GAAAAGGTAGTGGTTTGCGTG	Analysis of qRT-PCR or RT-PCR
	MG-2944	Reverse	CTTTGCTCTGTTTGCTTCCTG	
<i>GmCAMTA2</i> (<i>Glyma.08G135200</i>)	MG-2931	Forward	AAGGCTGGAAGTGTTGATG	
	MG-2932	Reverse	TTGGTTCCCTTCACTTGTCG	
<i>GmCAMTA3</i> (<i>Glyma.15G053600</i>)	MG-2957	Forward	AGGGACGGAACAAAAGGAAG	
	MG-2958	Reverse	ACCCTTGTTAGATGCCTTGG	
<i>GmCAMTA4</i> (<i>Glyma.08G072100</i>)	MG-2947	Forward	GTTCAGTATCCAGAGGCACG	
	MG-2948	Reverse	TCCCTTGGTTTGACGGAAG	

<i>GmCAMTA5</i> (<i>Glyma.05G117000</i>)	MG-2963	Forward	GTTTCAGTATCCAGAGGCACG	
	MG-2964	Reverse	CACCATCAACTGTCTCTTCCG	
<i>GmCAMTA6</i> (<i>Glyma.08G178900</i>)	MG-2951	Forward	AGGGACGGAACAAAAGGAAG	
	MG-2952	Reverse	AACCTCGTTAGATGCCTTG	
<i>GmCAMTA7</i> (<i>Glyma.17G038800</i>)	MG-2959	Forward	CAGATCCACCTGTTATGCCTC	
	MG-2960	Reverse	GCTTCTCTTACAGTCTTCCCATC	
<i>GmCAMTA8</i> (<i>Glyma.15G143400</i>)	MG-2933	Forward	CTGCGTTGGCGTTAAAGAG	
	MG-2934	Reverse	AATCACTTTCCTGCGTCTCC	
<i>GmCAMTA9</i> (<i>Glyma.09G038300</i>)	MG-2953	Forward	GTAGTCCTTTATCTTCGGGTGG	
	MG-2954	Reverse	GTTTCATCCATGTTCCAAAGCTG	
<i>GmCAMTA10</i> (<i>Glyma.05G148300</i>)	MG-2941	Forward	TGACATCAATGGGTGGACTG	
	MG-2942	Reverse	GGTTGGATCTTGTGCATTGG	
<i>GmCAMTA11</i> (<i>Glyma.18G005100</i>)	MG-2961	Forward	TGGGCAGTTGGAATCTTGG	
	MG-2962	Reverse	CGGAACACGCTGAGAAAATC	
<i>GmCAMTA12</i> (<i>Glyma.17G031900</i>)	MG-2935	Forward	CTCGCATACAGTGTACATACCG	
	MG-2936	Reverse	ACGACACCAACAGACCAAAG	
<i>GmCAMTA13</i> (<i>Glyma.07G242000</i>)	MG-2945	Forward	TCTTTGGTCTGTTGGTGTGG	
	MG-2946	Reverse	ATCGACAGTTTGACCTGGAG	
<i>GmCAMTA14</i> (<i>Glyma.11G251900</i>)	MG-2955	Forward	ATTCTCTGCCATGACTCGG	
	MG-2956	Reverse	ATGCCGTCCTGAATTATCTCTG	
<i>GmCAMTA15</i> (<i>Glyma.08G105200</i>)	MG-2949	Forward	GTCTTTTCTCTGTCATCCCTCG	
	MG-2950	Reverse	TGCACAGAGTAACCTTTCCAG	
<i>GmTUBULIN</i> (<i>Glyma.17G258300</i>)	MG-2937	Forward	TGGCCGTTACCTGACAGCAT	
	MG-2938	Reverse	CTCGGAGGGATGTCACACAC	
<i>AtRD29A</i> (<i>AT5G52310</i>)	MG-2000	Forward	CCTGAAGTGATCGATGCACCAG	
	MG-2001	Reverse	TGGTGTAATCGGAAGACACGAC	
<i>AtRD29B</i> (<i>AT5G52300</i>)	MG-2002	Forward	GTGAAGATGACTATCTCGGTGG	
	MG-2003	Reverse	CACCACTGAGATAATCCGATCC	
<i>AtP5CS2</i> (<i>AT3G55610</i>)	MG-2018	Forward	AGCAGCCTGTAATGCGATGG	
	MG-2019	Reverse	AAGTGACGCCTTTGGTTTGC	
<i>AtKIN1</i> (<i>AT5G15960</i>)	MG-2016	Forward	CCAACAAGAATGCCTTCCAAGC	
	MG-2017	Reverse	GCTGCCGCATCCGATACACT	
<i>AtTUBULIN2</i> (<i>AT5G62690</i>)	MG4028	Forward	TGGCATCAACTTTCATTGGA	
	MG-4029	Reverse	ATGTTGCTCTCCGCTTCTGT	
<i>GmCAMTA2</i> (<i>Glyma.08G135200</i>)	MG-1523	Forward	CCATGGCTGAGGCCAGACTCTAC	Gene cloning
	MG-1281	Reverse	CTCAGCTGTGAAATTCTAAATCACCCC	
<i>GmCAMTA8</i> (<i>Glyma.15G143400</i>)	MG-1530	Forward	CATCCCAAATATGGGGAAACCTTGTTT	
	MG-1531	Reverse	CATCCCAAATATGGGGAAACCTTGTTT	