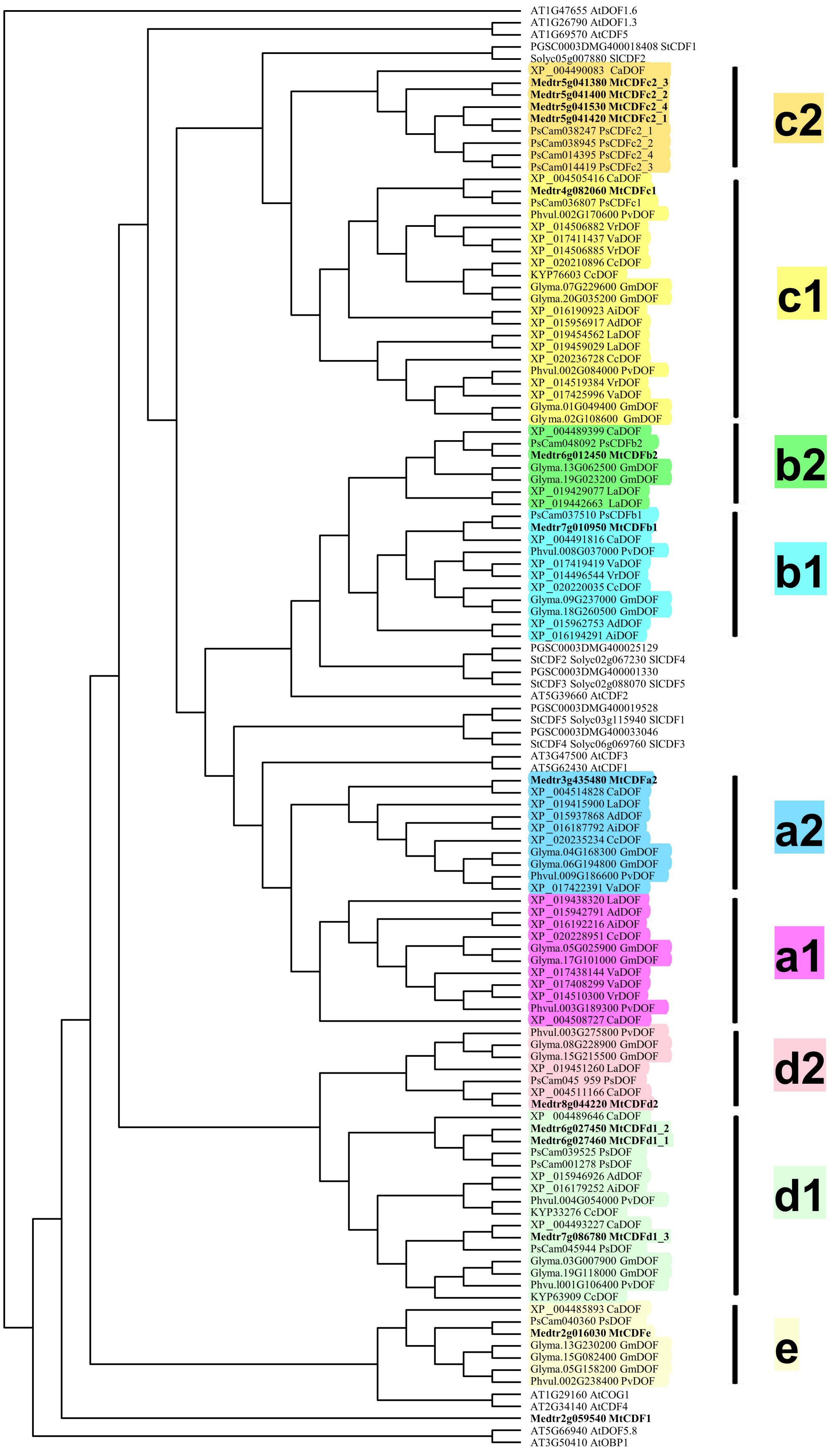
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Medtr5g041380 MtCDFc2_3
Medtr5g041400 MtCDFc2_2
Medtr5g041530 MtCDFc2_4
Medtr5g041420 MtCDFc2_1
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PsCam014395 PsCDFc2_4
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XP _017411437 VaDOF
XP _014506885 VrDOF
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Glyma.20G035200 GmDOF
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XP _015956917 AdDOF
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XP _020236728 CcDOF
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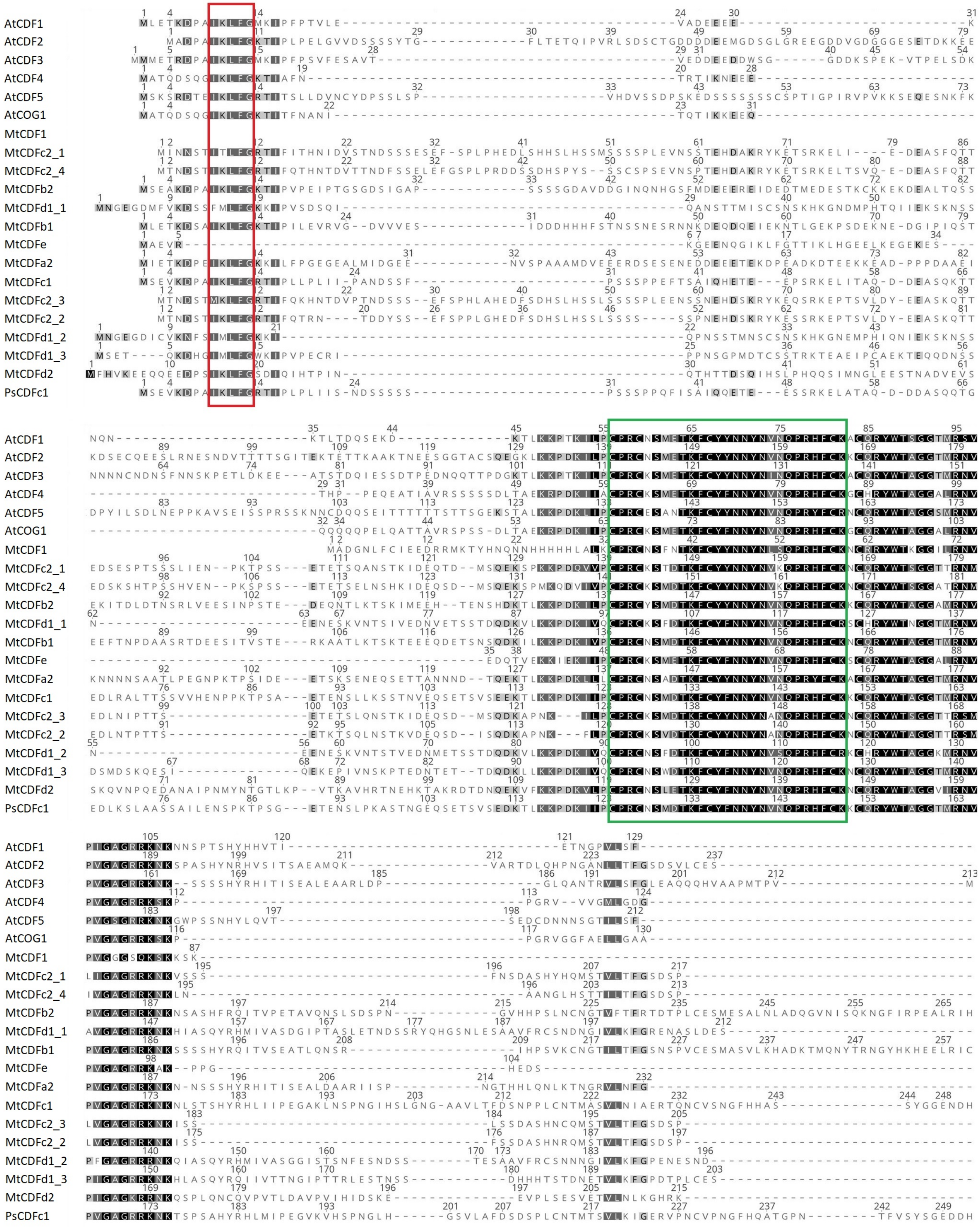
c2
c1
b2
b1
a2
a1
d2
d1
e

Supplementary Figure S1: Phylogenetic tree of CDF-like proteins from *Medicago,* other legumes, tomato, potato and Arabidopsis. The tree was constructed with full length amino acid sequences using the neighbour-joining algorithm implemented in PAUP* (version 4.0). Clades (a-c) of legume proteins have been named as described in Ridge et al. (2016). Additional nomenclature extended to clades d & e. Species included are; Ad - *Arachis duranensis,* Ai - *Arachis ipaensis,* At - *Arabidopsis thaliana, Ca - Cicer arietinum,* Cc - *Cajanus cajan,* Gm - *Glycine max,* La - *Lupinus augustifolius,* Mt - *Medicago truncatula,* Ps - *Pisum sativa.* Pv - *Phaseolus vulgaris,* Sl - *Solanum lycopersicum,* St - *Solanum tuberosum,* Va - *Vigna angularis* and Vr - *Vigna radiata.*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MtCDFb1
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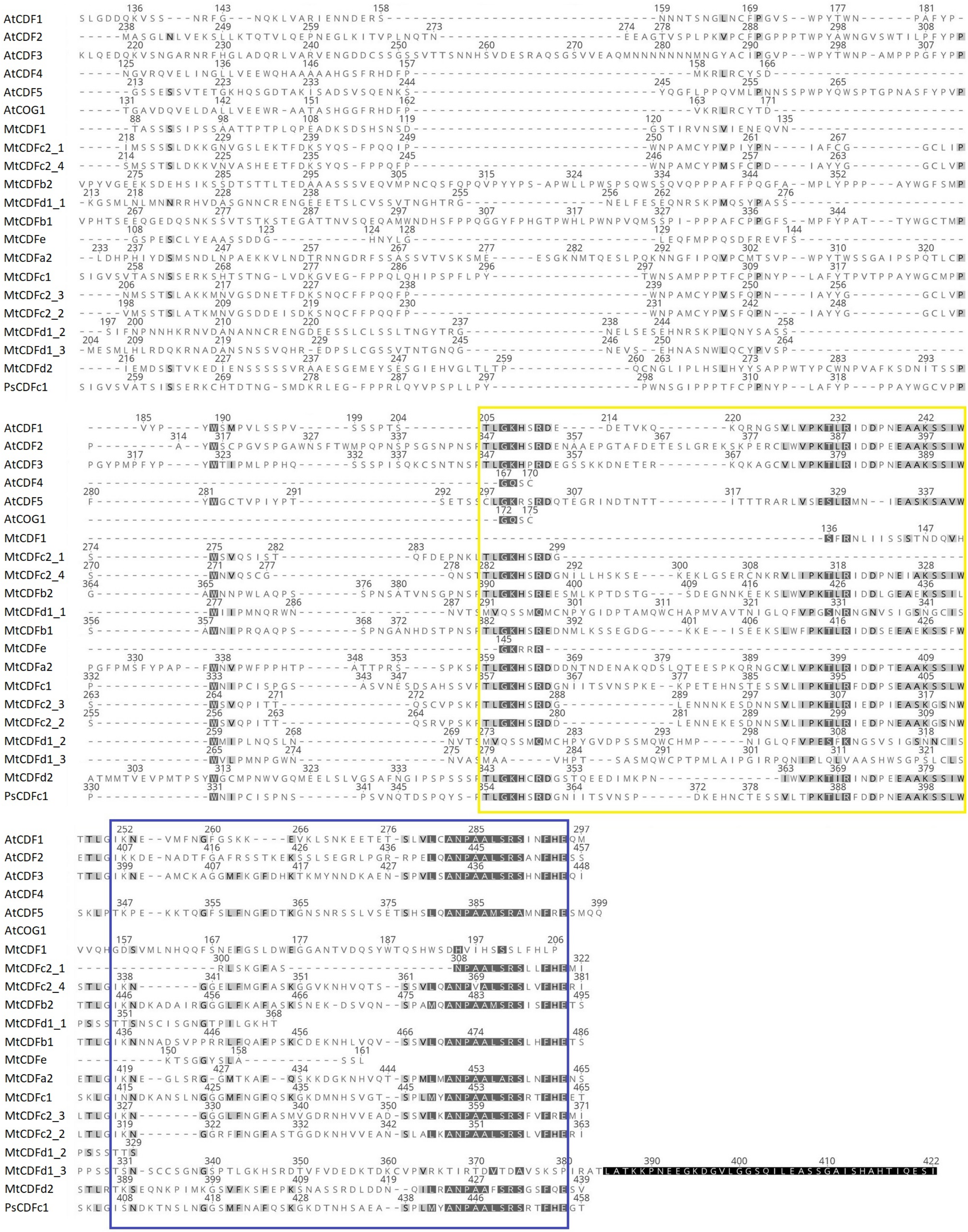
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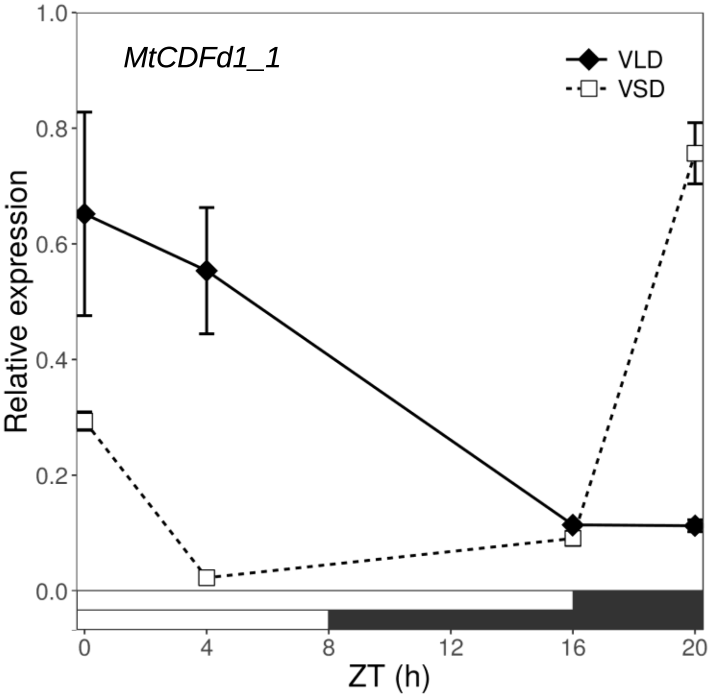
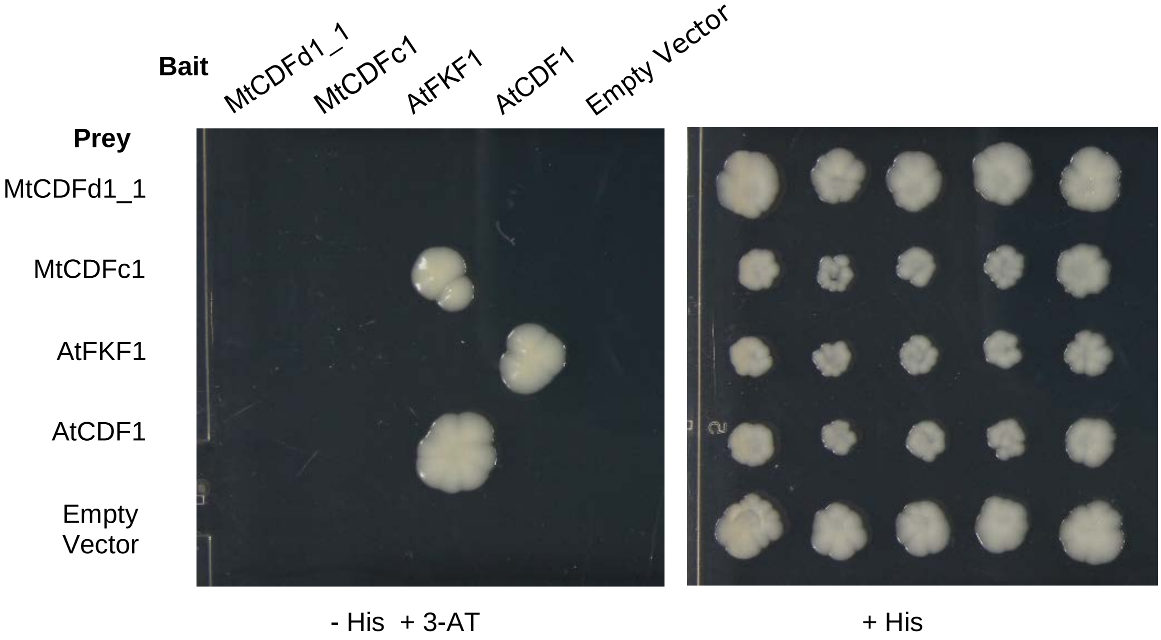
MtCDFb1
MtCDFe
MtCDFa2
MtCDFc1
MtCDFc2_3
MtCDFc2_2
MtCDFd1_2
MtCDFd1_3
MtCDFd2
PsCDFc1

Supplementary Figure S2: Alignment of Medicago, Arabidopsis and pea CDF protein sequences showing the highly conserved DOF domain (green box) present in all the proteins and three other domains present in most of the CDF proteins, the N- terminal TOPLESS (TPL) interacting domain (red box) and two C-terminal domains, a putative GI-interacting domain (yellow box) and a FKF1-interacting region (blue).



Supplementary Figure S3. *MtCDFd1_1* gene transcript levels have a diurnal cycle. Gene expression of *MtCDFd1_1* in a Jester diurnal time course in VSD and VLD. Results are presented as averages ± SE of three biological replicates at each time point and normalised to Medicago *PROTEIN PHOSPHATASE 2A* (*PP2A)*. Data are presented relative to the highest value observed overall. Bars underneath each graph show the length of light and dark conditions for LD (16 h light/8 h dark; upper) and SD (8 h light/16 dark; lower). ZT (zeitgeber time) indicates hours after dawn.

Each replicate consists of two fully-expanded trifoliate leaves from vernalised (V) wild type Jester plants grown in SD or LD.



Supplementary Figure S4. *MtCDFd1_1* does not interact with AtFKF1 in yeast two hybrid assays. (**B**) Diploid yeast colonies containing prey and bait constructs from *MtCDFd1_1*, *MtCDFc1*, AtCDF1 and FKF1 or empty vector controls, were grown on medium selecting for protein-protein interactions, lacking His with 2mM 3-AT (left) or on medium with His (right). Colonies were allowed to develop over 11 days at 28^{o}C. The vectors were pDEST22 (prey vector with the GAL4 activation domain) and pDEST32 (bait vector with GAL4 binding domain). Vectors containing inserts encoding full length CDF proteins were tested while the fragment encoding the KELCH-repeat region of AtFKF1 (amino acids 284 to 619) was used. The positive control interaction was between AtCDF1 and AtFKF1; AtCDF1, as either bait or prey, interacted with AtFKF1 (Imaizumi et al., 2005; Ridge et al., 2016). One of the Medicago CDFs, MtCDFc1 interacted with AtFKF1 only in one direction, when it was the prey. However, as predicted from the lack of conservation of the C-terminal domains with predicted GI and FKF1 binding functions, there was no evidence of interaction between MtCDFd1_1 and AtFKF1. Similar results were obtained with a second experiment using independent colonies.

Supplementary Table S1: Summary of the results of screening of *MtCDF Tnt1* insertion mutants.

Medicago gene ID[1]	**Gene Names[2]**	***Thomson et al.* Nomenclature[4]**	***Tnt1* line**	***Tnt1* position**	**Fl. time LD[+]**		**Fl. time VLD[+]**		**Fl. time VSD[+]**		**Gene expression KD/KO***
					Days	**Nodes**	**Days**	**Nodes**	**Days**	**Nodes**	
Medtr2g059540	*CDF1*[3]	*CDF1*	NF9016	exon 1	65.1±1.49	17.6±0.28	23.1±0.61	8.4±0.51	63.1±4.00	13.8±0.81	yes
Medtr5g041420	*CDFc2_1*	*CDF2*	NF13527	exon 2	62.4±0.75	17.1±0.61	25.9±0.77	9.1±0.36	63.1±1.42	13.3±0.57	yes
Medtr5g041530	*CDFc2_4*	*CDF3*	NF21008	exon 2	nd	nd	28.7±1.84	8.8±1.23	69.4±0.70	14.4±0.43	yes
			NF21606	exon 2	65.9±0.80	18.3±0.41	21.5±0.76	7.7±0.68	52.6±1.63	12.8±0.33	yes
Medtr6g012450	*CDFb2*	*CDF4*	NF0134	no Tnt1	na	na	na	na	na	na	na
			NF10121	no Tnt1	na	na	na	na	na	na	na
			NF11690	intron 1	61.8±0.87	16.8±0.36	24.4±1.17	8.6±0.44	nd	nd	no
Medtr6g027460	*CDFd1_1*[3]	*CDF5*	NF9161	no Tnt1	na	na	na	na	na	na	na
			NF9868	exon 2	nd	nd	19.0±1.15	6.0±0.00	nd	nd	yes
			NF20445	no Tnt1	na	na	na	na	na	na	na
Medtr7g010950	*CDFb1*	*CDF6*	NF6504	intron 1	nd	nd	20.5±6.35	7.0±0.00	nd	nd	no
			NF14323	intron 1	63.5±2.76	17.2±0.81	22.3±1.22	8.1±0.42	nd	nd	no
			NF13712	no Tnt1	na	na	na	na	na	na	na
Medtr2g016030	*CDFe*[3]	*CDFa*	NF7191	exon 1	58.7±2.63	16.4±0.54	28.3±1.59	9.9±0.43	69.5±0.77	14.4±0.63	yes
			NF16323	no Tnt1	na	na	na	na	na	na	na
Medtr3g435480	*CDFa2*	*CDFb*	NF9123	exon 2	66.1±2.14	18.2±0.56	27.9±0.83	10.5±0.34	68.0±0.99	13.6±0.46	yes
			NF11050	exon 2	nd	nd	30.2±2.50	11.1±0.83	nd	nd	yes
Medtr4g082060	*CDFc1*	*CDFc*	NF1963	no Tnt1	na	na	na	na	na	na	na
			NF14291	exon 2	55.3±2.42	17.2±0.67	23.6±1.30	8.5±0.70	65.5±0.78	14.0±0.40	yes
Medtr5g041380	*CDFc2_3*	*CDFd*	NF2921	no Tnt1	na	na	na	na	na	na	na
			NF4354	exon 1	64.1±2.31	16.4±0.80	27.7±0.89	10.4±0.39	59.5±0.98	12.9±0.38	yes
Medtr5g041400	*CDFc2_2*	*CDFe*	NF11400	exon 1	59.9±3.13	16.3±0.64	28.7±1.59	10.7±0.83	60.2±0.82	13.9±0.47	yes
			NF17470	exon 2	nd	nd	21.6±0.73	7.4±0.60	nd	nd	yes
Medtr6g027450	*CDFd1_2*[3]	*CDFf*	NF1515	exon 2	nd	nd	20.1±0.98	7.2±0.64	nd	nd	nd
			NF19175	exon 2	63.4±4.66	17.5±1.61	22.3±1.07	7.8±0.30	61.1±5.08	13.6±0.73	yes
Medtr7g086780	*CDFd1_3*[3]	*CDFg*	NF11642	exon 2	57.3±1.99	16.1±0.46	24.1±0.45	8.8±0.32	59.9±0.77	13.9±0.34	yes
			NF21417	exon 2	nd	nd	20.9±1.36	6.8±0.64	nd	nd	nd
Medtr8g044220	*CDFd2*[3]	*CDFh*	NF16568	no Tnt1	na	na	na	na	na	na	na
R108 WT					58.0±0.78[a]	15.5±0.34[a]	23.9±0.59[c]	8.5±0.30[c]	58.7±1.00[g]	13.0±0.36[g]	
					54.4±0.75[b]	16.0±0.26[b]	22.7±0.96[d]	7.6±0.48[d]	69.5±0.58[h]	13.4±0.40[h]	
							26.1±0.65[e]	8.6±0.40[e]	63.3±1.34[i]	12.9±0.42[i]	
							24.5±0.68[f]	8.2±0.42[f]			

Fl., flowering; WT, wild type; nd, not done; na, not applicable; KD/KO, knock-down/knock-out

[1] Medicago gene identifiers from JCVI Medicago genome Mt4.0, http://www.jcvi.org/medicago/

[2] Gene names based on nomenclature from Ridge et al. 2016; [3] Additional extended nomenclature based on clades seen in Supplementary Figure 1

[4] Nomenclature from Thomson et al. 2019

* Based on qRT-PCR and/or full length cDNA amplification

[+] Values expressed as means ± 95% confidence intervals

[a] Same batch as NF9016, NF13527, NF21606, NF11690 and NF14323 in LD; [b] same batch as NF7191, NF9123, NF14291, NF4354, NF11400, NF19175 and NF11642 in LD; [c] same batch as NF9016, NF13527, NF21008, NF21606, NF11690 and NF14323 in VLD; [d] same batch as NF9868 and NF6504; [e] same batch as NF7191, NF9123, NF11050, NF14291, NF4354, NF11400, NF17470 and NF11642 in VLD; [f] same batch as NF1515, NF19175 and NF21417 in VLD; [g] same batch as NF9016, NF13527, NF21008 and NF21606 in VSD; [h] same batch as NF7191, NF9123 and NF14291 in VSD; [i] same batch as NF4354, NF11400, NF19175 and NF11642 in VSD

Supplementary Table S2: Primers used in this study

Primer name	**Primer sequence**	**Primer used for**
GW_Medtr2g059540-F (MtCDF1)	GGGGACAAGTTTGTACAAAAAAGCAGGCTATGGCAGATGGAAATCTTTTCTG	Gateway cloning
GW_Medtr2g059540-R (MtCDF1)	GGGGACCACTTTGTACAAGAAAGCTGGGTTCATGGAAGATGAAAAAGAGAGGA	
GW_Medtr5g041420-F (MtCDFc2_1)	GGGGACAAGTTTGTACAAAAAAGCAGGCTATGATTAATAATTCAACCATAACGCTT	
GW_Medtr5g041420-R (MtCDFc2_1)	GGGGACCACTTTGTACAAGAAAGCTGGGTGCCTTCAAACCAAGGTGTC	
GW_Medtr5g041530-F (MtCDFc2_4)	GGGGACAAGTTTGTACAAAAAAGCAGGCTAGCAAAAACCTACCTGAGATGACT	
GW_Medtr5g041530-R (MtCDFc2_4)	GGGGACCACTTTGTACAAGAAAGCTGGGTACACCGGACACACCTTCAAA	
GW_Medtr6g012450-F (MtCDFb2)	GGGGACAAGTTTGTACAAAAAAGCAGGCTATGTCTGAAGCAAAAGACCCTG	
GW_Medtr6g012450-R (MtCDFb2)	GGGGACCACTTTGTACAAGAAAGCTGGGTTGCTGTGTCAATCGGCTCAT	
GW_Medtr6g027460-F (MtCDFd1_1)	GGGGACAAGTTTGTACAAAAAAGCAGGCTCACAAAAATGAATGGTGAAGGTGAC	
GW_Medtr6g027460-R (MtCDFd1_1)	GGGGACCACTTTGTACAAGAAAGCTGGGTACAGAATCTCATGTGTGTTTGCC	
GW_Medtr7g010950-F (MtCDFb1)	GGGGACAAGTTTGTACAAAAAAGCAGGCTCTGTTTTTAGCAACAAAAATGTTGGA	
GW_Medtr7g010950-R (MtCDFb1)	GGGGACCACTTTGTACAAGAAAGCTGGGTGGTTGAACACGGCCAAATCA	
GW_Medtr2g016030-F (MtCDFe)	GGGGACAAGTTTGTACAAAAAAGCAGGCTAATATGGCTGAAGTGAGAAAAGGTG	
GW_Medtr2g016030-R (MtCDFe)	GGGGACCACTTTGTACAAGAAAGCTGGGTACAAGAAATCACAAGGAGGATGCTA	
GW_Medtr3g435480-F (MtCDFa2)	GGGGACAAGTTTGTACAAAAAAGCAGGCTAAAAATGATTGAAACTAAGGACCCT	
GW_Medtr3g435480-R (MtCDFa2)	GGGGACCACTTTGTACAAGAAAGCTGGGTTGACTAACCTCAATTCTCAAACACA	
GW_Medtr4g082060-F (MtCDFc1)	GGGGACAAGTTTGTACAAAAAAGCAGGCTTGAGTTGCACACAGATTTTCCT	
GW_Medtr4g082060-R (MtCDFc1)	GGGGACCACTTTGTACAAGAAAGCTGGGTTGCAATTCACAACTAGGTTTCCT	
GW_Medtr6g027450-F (MtCDFc1_2)	GGGGACAAGTTTGTACAAAAAAGCAGGCTTGGTGTGTTTTTGAGCTGTT	
GW_Medtr6g027450-R (MtCDFc1_2)	GGGGACCACTTTGTACAAGAAAGCTGGGTATTACCGGAGCGACAACTAT	
GW_Medtr7g086780-F (MtCDFd1_3)	GGGGACAAGTTTGTACAAAAAAGCAGGCTATGTCTGAGACTCAGAAGGATC	
GW_Medtr7g086780-R (MtCDFd1_3)	GGGGACCACTTTGTACAAGAAAGCTGGGTTTATATGCTTTCCTGGATTGTATG	
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Medtr5g041420-F (MtCDFc2_1)	ACCATCTTGGAGCGTGCAAT	
Medtr5g041420-R (MtCDFc2_1)	AGCTGCAGGGTTAGAAGCAA	
Medtr5g041530-F (MtCDFc2_4)	AAGCGCGTGTTGATCCCTAA	
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R108_Medtr6g027460-R (MtCDFd1_1)	TGGACACATTTCCATTCCTG	
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Medtr2g016030-R (MtCDFe)	CACATGGGAGGAAAATGGTT	
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Medtr3g435480-R (MtCDFa2)	CATTTTCGTCGTTGGTGTTG	

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Medtr5g041400-R (MtCDFc2_2)	GGACGTTGAATCGACAGGAC	
Medtr6g027450-F (MtCDFc1_2)	GCCGAACATTGGCTTGCAAT	
Medtr6g027450-R (MtCDFc1_2)	TCTCTGGTGTGTTTTCCAAGG	
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Medtr7g086780-R (MtCDFd1_3)	AGAGAAACAAAGGCCAGCAA	
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MtFTa1-R	ACACTCACTCTCGGTTGATTTCC	
MtFTb1-F	ATGAACCCTCTTGTGGTCTG	
MtFTb1-R	TGGATTGACTATTTGGGAAG	
MtFTb2-F	ACAAATCCTCTTGTTGTTGG	
MtFTb2-R	TGAGTTGATTATTTGAGAGG	
MtFTa2-F	AAGTGGTAGCAGACCGAATC	
MtFTa2-R	CACCACCATTGGTAACAGTC	
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MtCOLa-F	GGTTGTGAGGTGGAGGAAGA	
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MtCOLb-F	CCGAACACGAACATGAACAC	
MtCOLb-R	CATCCATCGACGGTGAAGA	
MtCOLc-F	ATCTTCCTCGATGGACGTTG	
MtCOLc-R	GTCCCGTAACCACAGTTTGA	
MtCOLd-F	GCCGATGATGAATCACAACTCAG	
MtCOLd-R	GTGAAGATGACGAAACGCTATG	
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MtCOLh-R	CAAATCGCCCTCTCACTCTC	
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Medtr5g041420_genotyping-R (MtCDFc2_1)	TGGAACAACTTGGTCTGGTTTC	
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Medtr6g012450_genotyping-R2 (MtCDFb2)	AATCGCATCTGCCTTATCGT	
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Medtr6g027460_genotyping-R (MtCDFd1_1)	TGGACACATTTCCATTCCTG	
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Medtr6g027460-R (MtCDFd1_1)	GCATTGCAGTAGGATCAATGCC	
Medtr7g010950_genotyping-F (MtCDFb1)	TCGTGTTGGAGATGTTGTTGT	
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Medtr7g010950_genotyping-F2 (MtCDFb1)	AGTGAAGAACAAGGCGAGGA	
Medtr7g010950_genotyping-R (MtCDFb1)	CCGATGGTTTTTCTCCCAATGT	
Medtr7g010950_genotyping-R1 (MtCDFb1)	CGGAATCACGTTGAGTTTCA	
Medtr7g010950_genotyping-R2 (MtCDFb1)	TGAAGCGACCTAGACAATGC	
Medtr2g016030_genotyping-F (MtCDFe)	CGGAAAGTTGTCTCTACGAAGC	
Medtr2g016030_genotyping-R (MtCDFe)	TGAATAACCACCTGAGGTCTTCC	
Medtr3g435480_genotyping-F (MtCDFa2)	TGGTAGAGTTCTCAACTTTGGGT	
Medtr3g435480_genotyping-R (MtCDFa2)	ATTGCGCCAGAACTCCAAGT	
Medtr4g082060_genotyping-F (MtCDFc1)	ACTGATCATCCCTGCCAACG	
Medtr4g082060_genotyping-R (MtCDFc1)	ACGAGGCTGGTTGACATTGT	
Medtr5g041380_genotyping-F (MtCDFc2_3)	ACAACACCGATGTTCCTACCA	
Medtr5g041380_genotyping-R (MtCDFc2_3)	CCTCTGTGGTTTGTTTTGAGGC	
Medtr5g041400_genotyping-F (MtCDFc2_2)	ACACGCAACACCGACGATTA	
Medtr5g041400_genotyping-R (MtCDFc2_2)	GAGATTTTGTTCTTGCGGCG	
Medtr6g027450_genotyping-F (MtCDFc1_2)	CCCGACAAAATTGTTCAATGTCC	
Medtr6g027450_genotyping-R (MtCDFc1_2)	GGTTGAAATTCCACCGGAAGC	
Medtr7g086780_genotyping-F (MtCDFd1_3)	CGATGCAACAGTTGGGACAC	
Medtr7g086780_genotyping-R (MtCDFd1_3)	TCGAACCGGAACACACTTGT	
Medtr8g044220_genotyping-F (MtCDFd2)	GATGCAGTACCTGTCATACACAT	
Medtr8g044220_genotyping-R (MtCDFd2)	AGAAGAGCTGGAAGAGTTCTCA	
Tnt1-F	ACAGTGCTACCTCCTCTGGATG	
Tnt1-R	CAGTGAACGAGCAGAACCTGTG	

Supplementary References

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