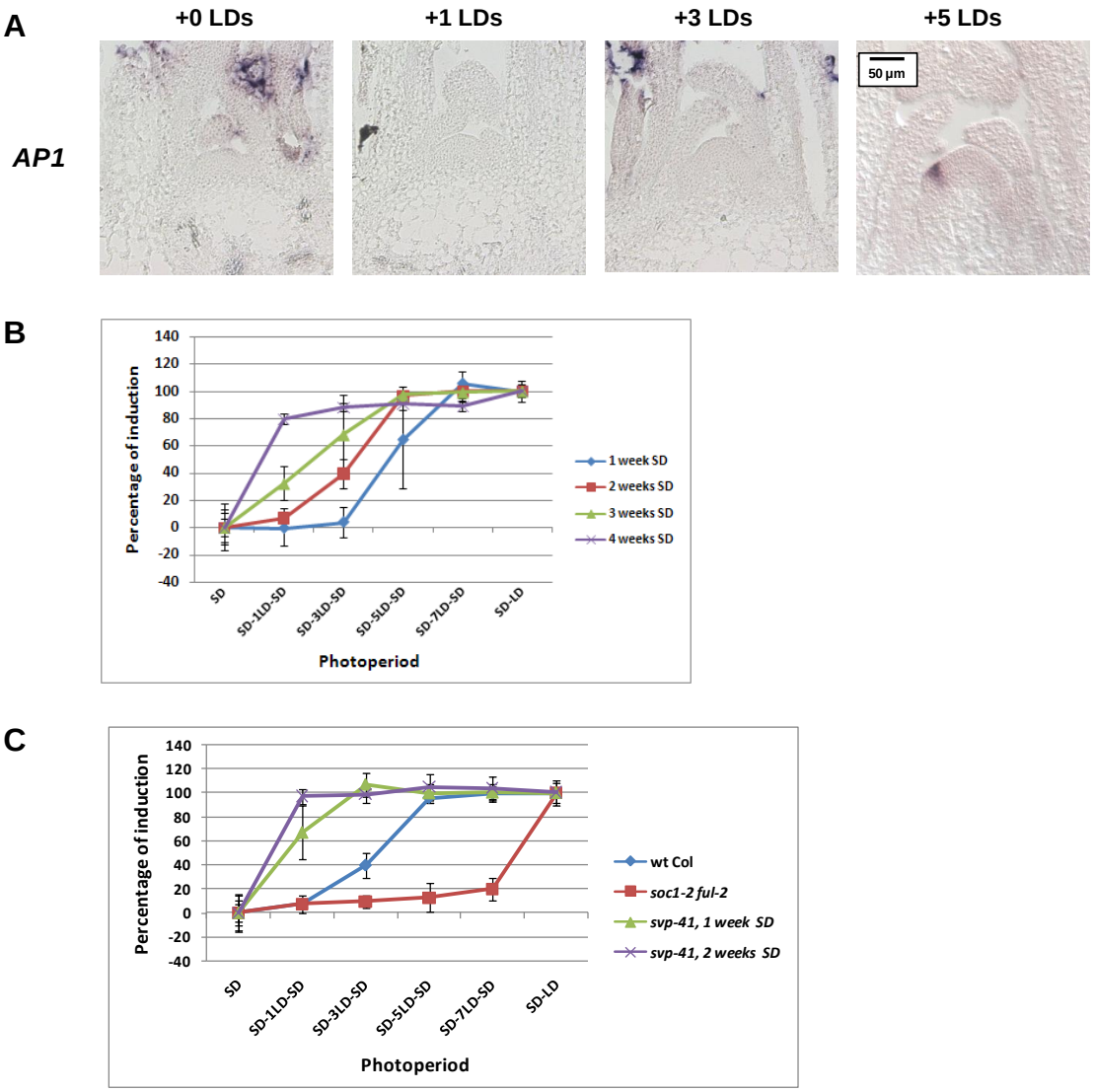


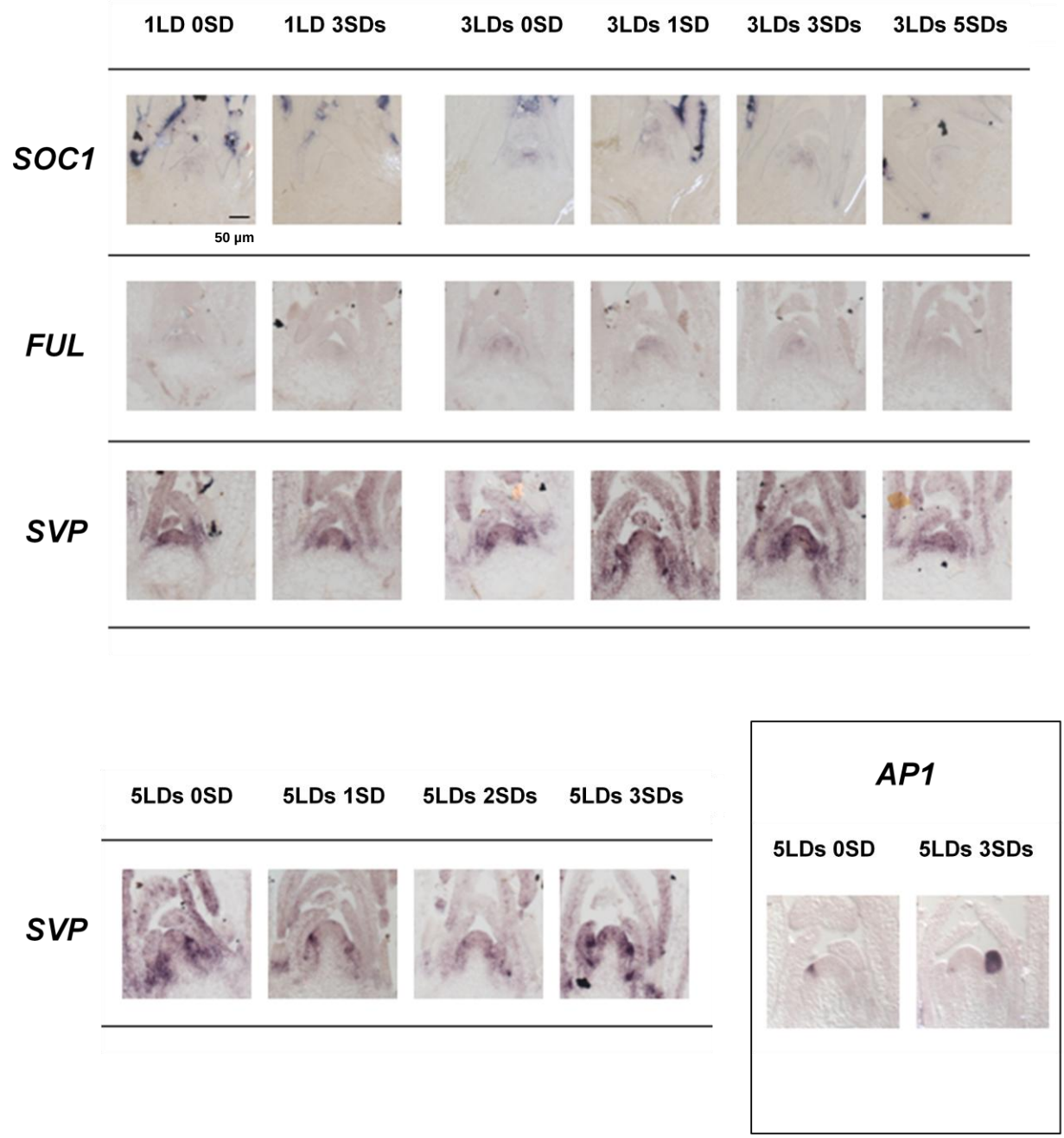
Supplemental Figure 1. Floral commitment in *Arabidopsis* WT and mutants.

(A) *In situ* hybridization of *APETALA1* probe on WT Col grown for two weeks in SDs and shifted to LDs for the indicated number of days.

(B) Commitment of WT Col to flower with different duration of the initial growth in SDs.

(C) Commitment of plant with different genotypes with initial growth of 2 weeks in SDs.

In **(B)** and **(C)**: Exposure to day length as shown on X-axis. Y-axis, percentage of induction, calculated as described in the Methods. Error bars represent standard deviation. At least eight plants were used to score flowering time of each condition or genotype.

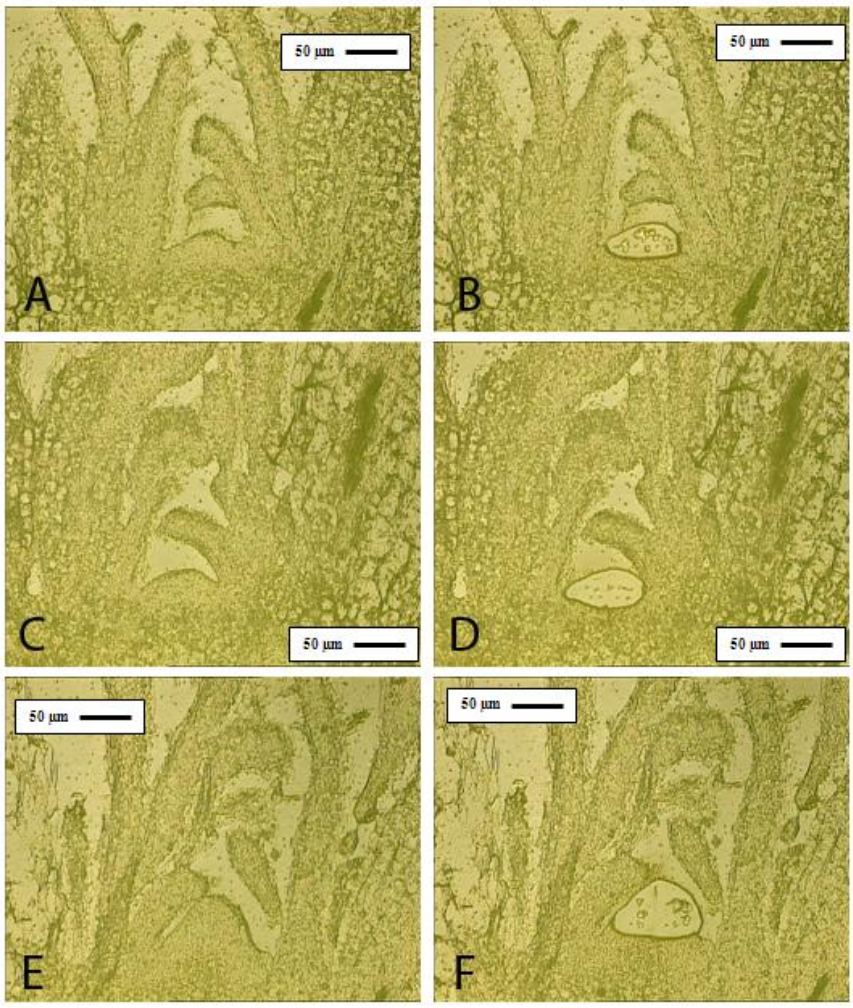


Supplemental Figure 2. *In situ* hybridization on WT Col apices in double shift experiments using probes for *SOC1*, *FUL*, *SVP* and *AP1*.

Analysis of gene expression after transient exposure of SD-grown plants to LDs. Plants were grown for two weeks in SDs, transferred to LDs and transferred back to SDs as indicated above each panel. Scale bar is 50 µm and refers to all panels.

Supplemental Figure 3. Comparison of inflorescences of *soc1-2 ful-2* and *svp-41 soc1-2 ful-2* plants.

Photographs were taken after flowering and represent plants sown at the same time and grown in the same conditions.



Supplemental Figure 4. Laser microdissection performed on WT Col apices during the floral transition.

Plants were grown for two weeks in SDs prior to LD induction.

(A) 0 LD, before microdissection.

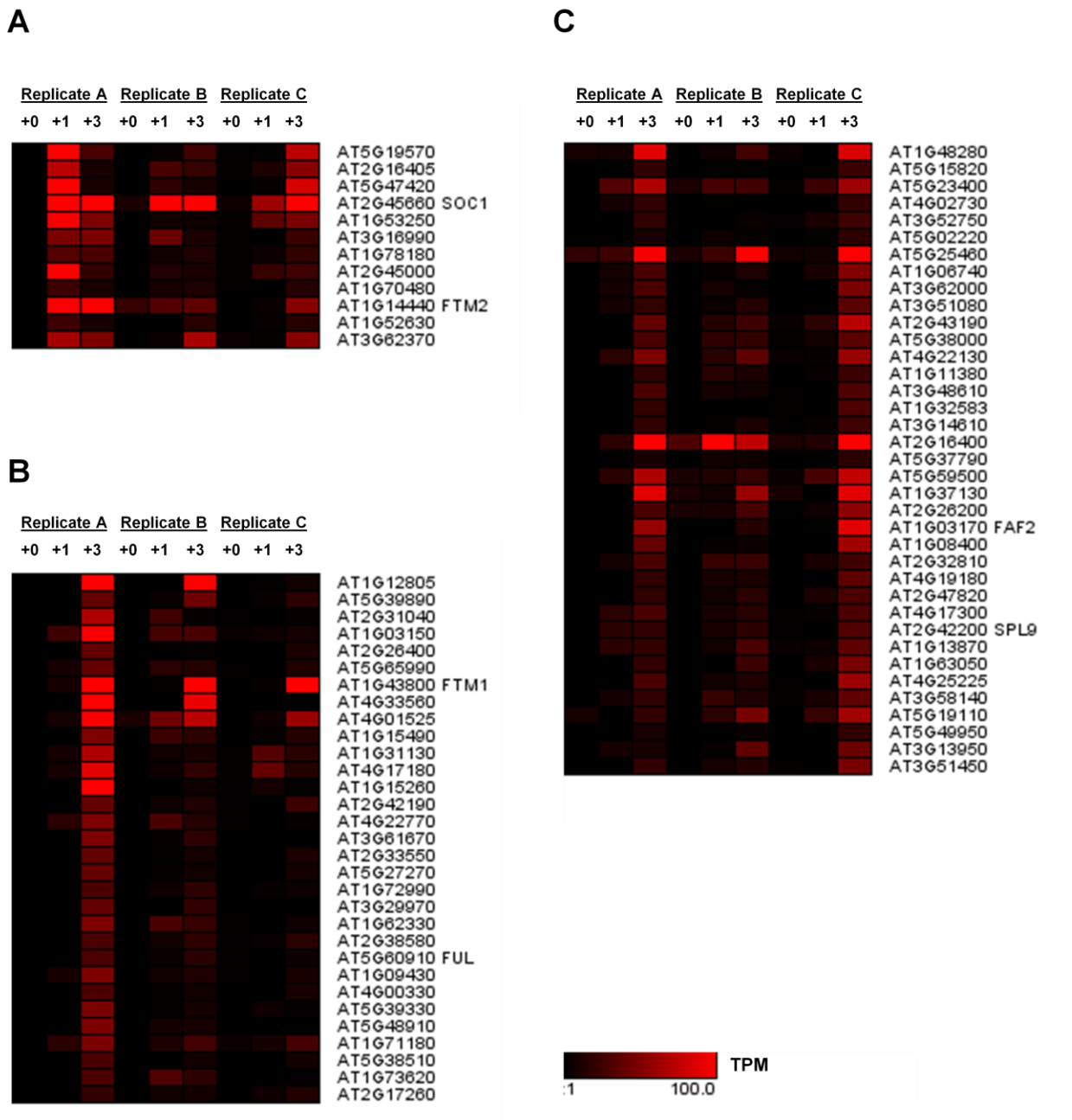
(B) 0 LD, after microdissection.

(C) 1 LD, before microdissection.

(D) 1 LD, after microdissection.

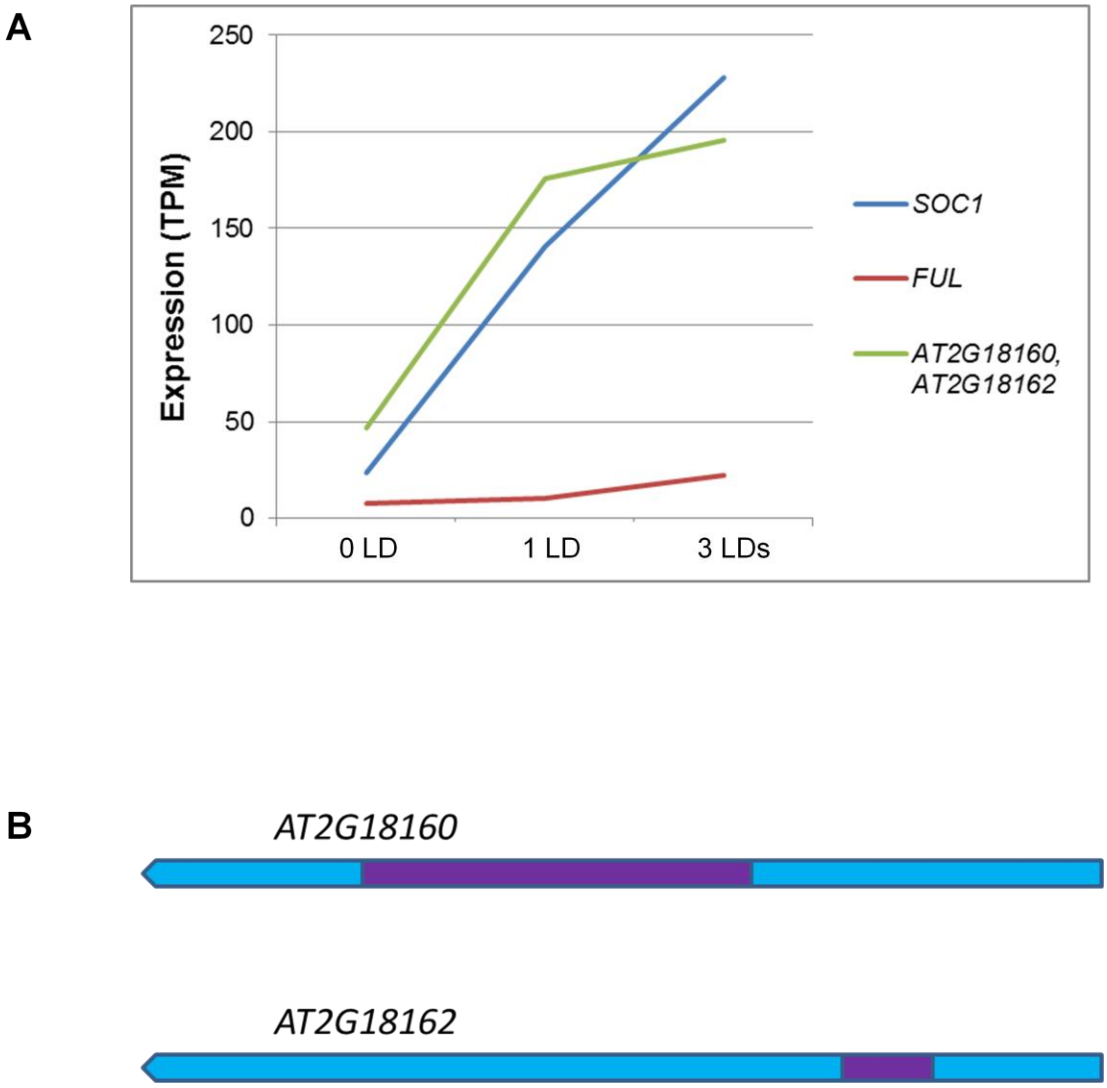
(E) 3 LDs, before microdissection.

(F) 3 LDs, after microdissection.



Supplemental Figure 5. K-means clustering of the up-regulated genes.

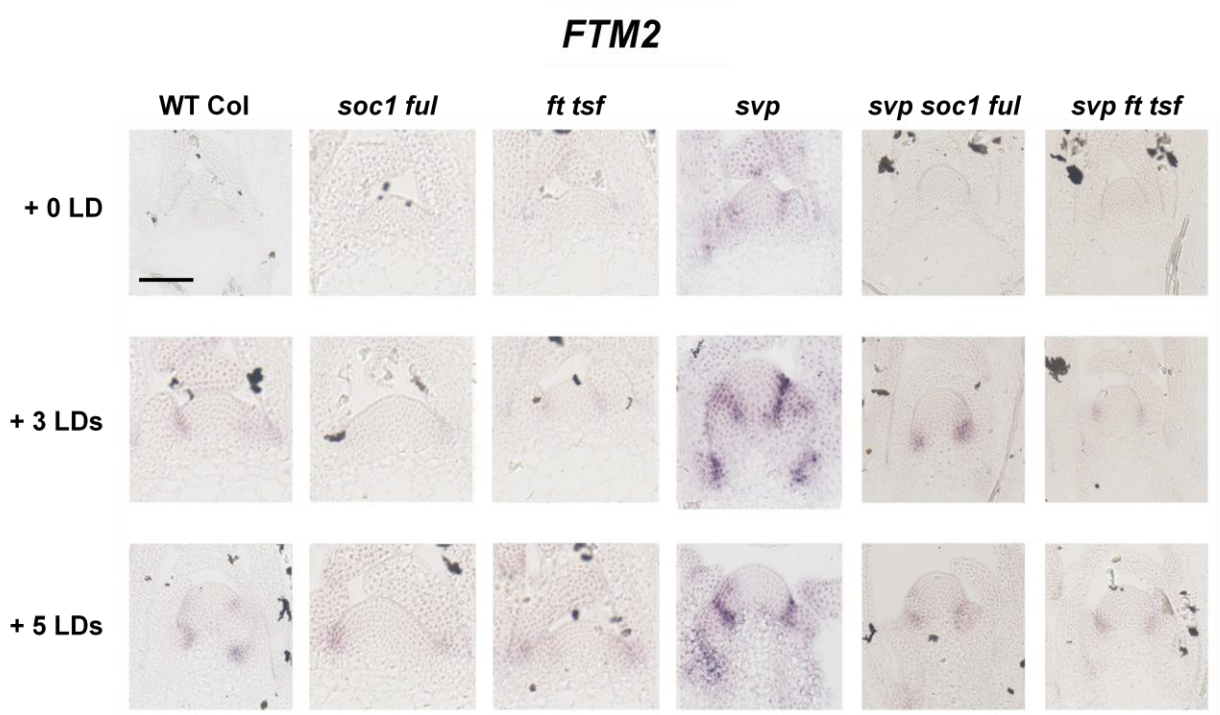
Genesis software was used to clusters the 202 up-regulated genes. Three (panels **(A)**, **(B)** and **(C)**) of the 8 clusters generated are shown in the figure. K-means clustering was performed using “Pearson squared” as distance. TPM: transcripts per million (see text for details). For each replicate, numbers represent number of long days after shift.

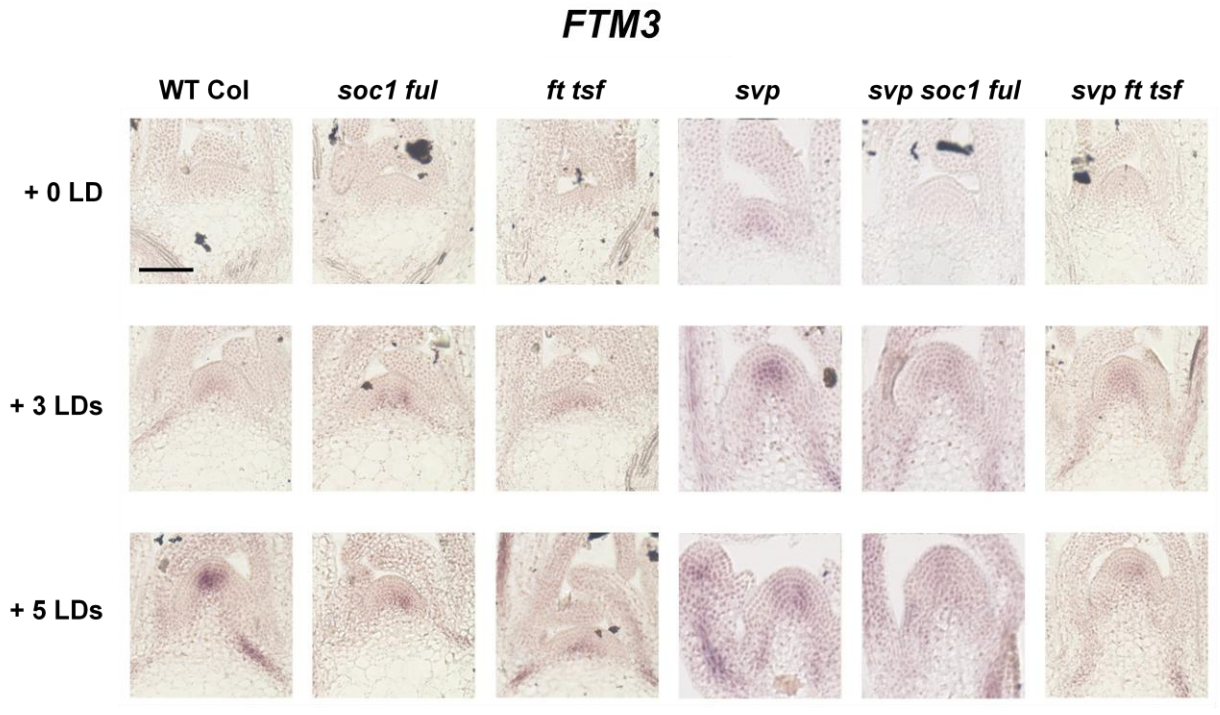


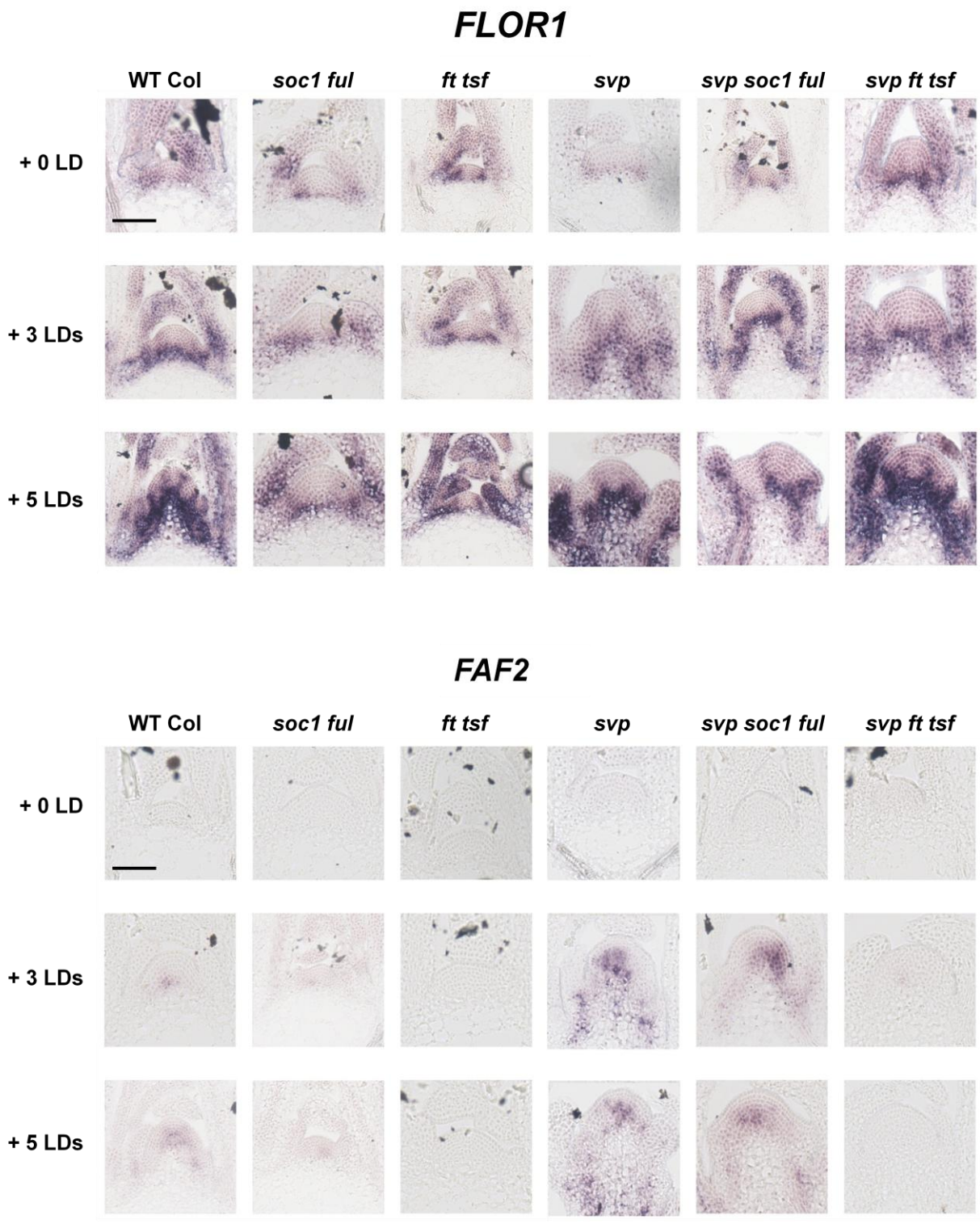
Supplemental Figure 6. Annotation and sequence reads of *FTM3* (At2G18160).

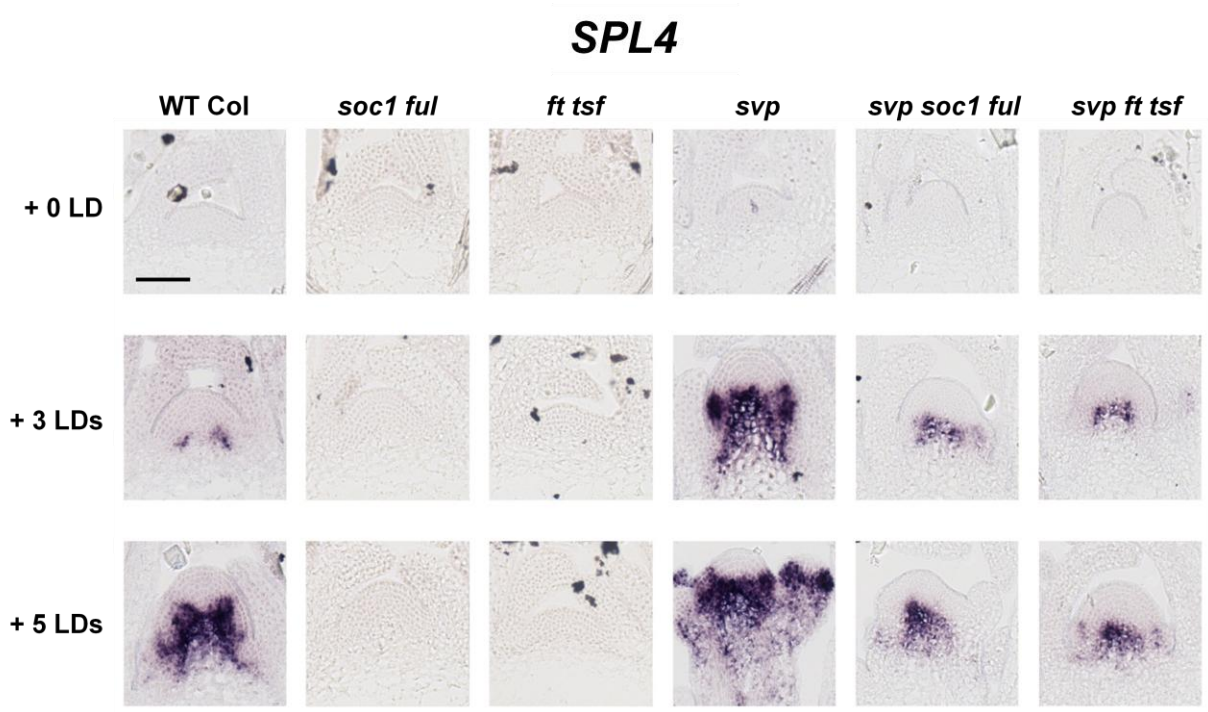
(A) Number of reads of both TAIR annotated ORFs combined show induction similar to that observed for *SOC1*. TPM: transcripts per million (see text for details).

(B) Schematic representation of double annotation of At2G18160 and At2G18162 in TAIR (which are actually both part of At2G18160).


FTM2
WT Col
soc1 ful
ft tsf
svp
svp soc1 ful
svp ft tsf
+ 0 LD
+ 3 LDs
+ 5 LDs

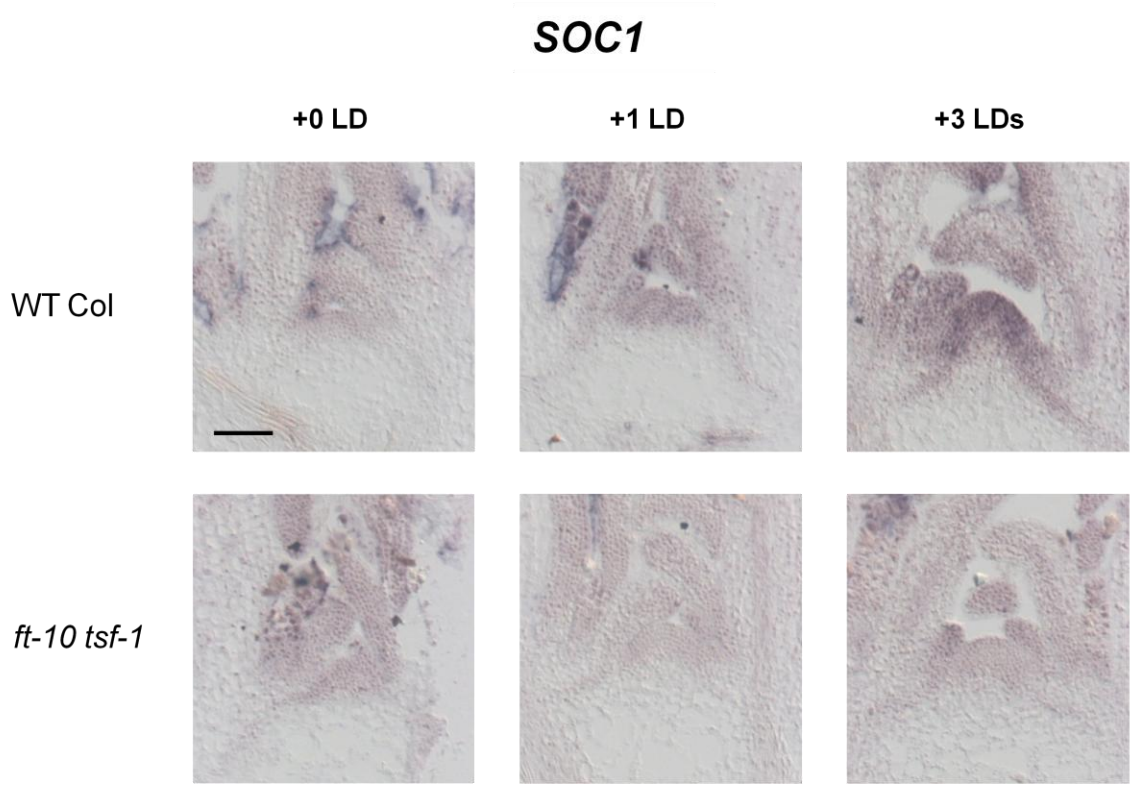

FTM3
WT Col
soc1 ful
ft tsf
svp
svp soc1 ful
svp ft tsf
+ 0 LD
+ 3 LDs
+ 5 LDs


FLOR1
WT Col
soc1 ful
ft tsf
svp
svp soc1 ful
svp ft tsf
+ 0 LD
+ 3 LDs
+ 5 LDs
FAF2
WT Col
soc1 ful
ft tsf
svp
svp soc1 ful
svp ft tsf
+ 0 LD
+ 3 LDs
+ 5 LDs



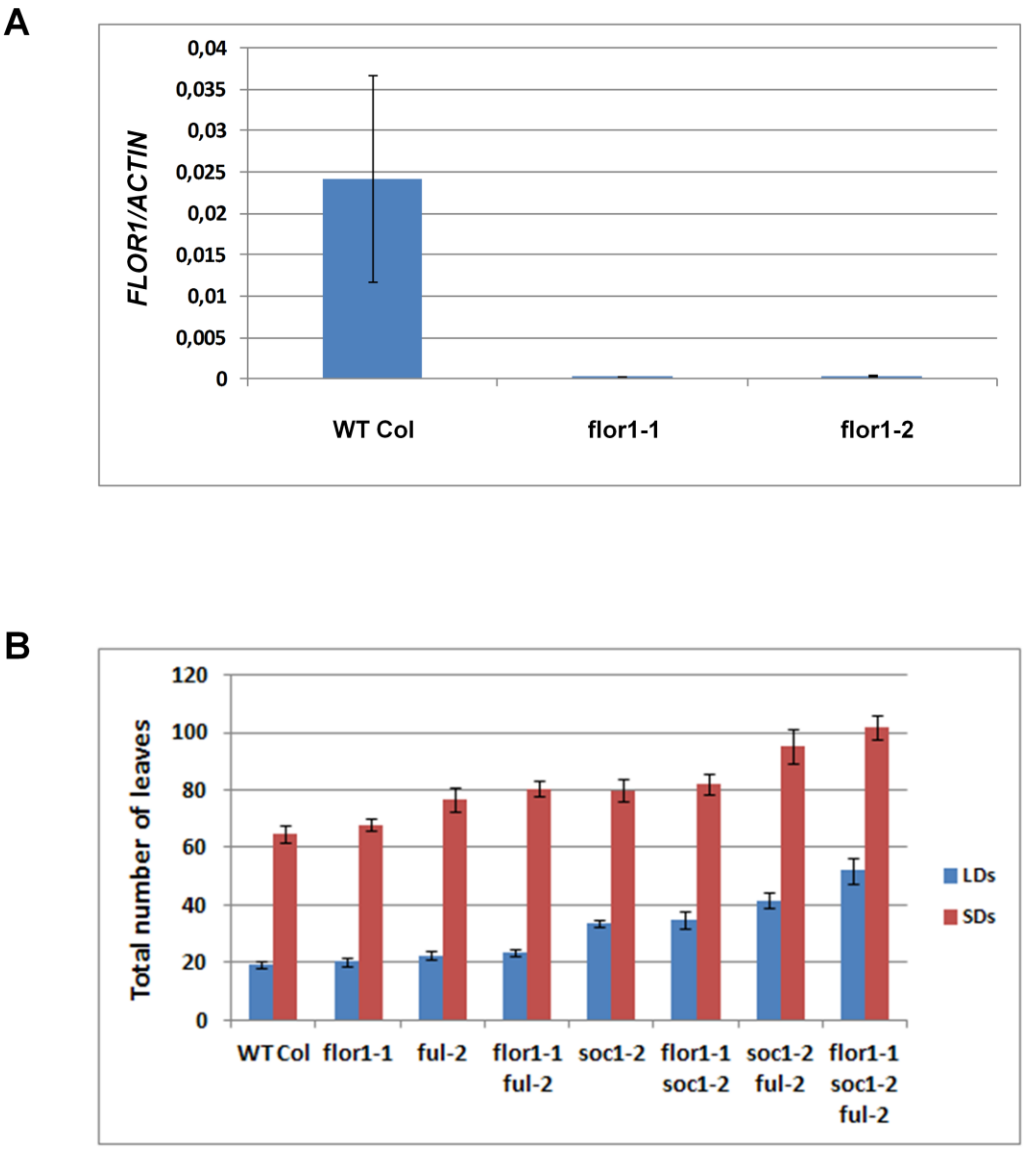
Supplemental Figure 7. Time course of *in situ* hybridizations in different genotypes for genes identified as being upregulated during floral induction.

Complete *in situ* hybridization time courses of on apices of WT Col and the illustrated mutant genotypes grown for two weeks in SDs and then transferred to LDs for 3 and 5 LDs. The genes used as probes are shown on the top of each panel. The *svp-41* mutant flowers early under SDs, and therefore these plants were grown for only 10 SDs before transfer. Samples were harvested at ZT8. Scale bar is 50 μm.



Supplemental Figure 8. Time course analysis of *SOC1* expression in Col and *ft-10 tsf-1.*

SOC1 expression upon transfer from SDs to LDs in WT Col and *ft-10 tsf-1*. Scale bar is 50 µm.



Supplemental Figure 9. Genetic and molecular analysis of *flor1* mutations.

(A) Expression of *FLOR1* measured by q-RT PCR. The two lines with T-DNA insertions are compared to WT Col. Two biological replicates of the experiment were performed and for each of them three technical replicates were compared in the PCR reaction. Error bars represent standard deviation in one experiment.

(B) Flowering time scored as total number of leaves both in LD and SD conditions for *flor1-1* allele in combination with other mutations and control lines. At least eight genetically identical plants were used to score flowering time of each genotype. Error bars represent standard deviation.

Supplemental Table 1. List of primers used in this study.

Genotyping		
soc1-2	SOC1-F	TTCTTCTCCCTCCAGTAATGC
	SOC1-R	GAGTTTTGCCCCTCACCATA
	SALKLB-R	TGGTTCACGTAGTGGGCCATCG
svp-41	SVP-1F	GACCCACTAGTTATCAGCTCAG
	SVP-1R (wt)	AAGTTATGCCTCTCTAGGAC
	SVP-2R (mut)	AAGTTATGCCTCTCTAGGTT
ful-2	FULseq2F	GCCGAGACGTTTCACAAAGT
	FULseq2R	TTGCCTTGAGCTTAGCATGT
flor1-1 and *flor1-2*	FLOR1-F3	GCATCCACACATAATCACGA
	FLOR1-R	AAGGGGAGTTCCACAAAGAC
	SALKLB-F	TGGTTCACGTAGTGGGCCATCG
Real-time PCR		
ACTIN2	qRT-ACT-F	GGTAACATTGTGCTCAGTGGTGG
	qRT-ACT-R	AACGACCTTAATCTTCATGCTGC
FT	FT-F	CGAGTAACGAACGGTGATGA
	FT-R	CGCATCACACACTATATAAGTAAAACA
SOC1	SOC1small-R	GTGATCTCCACTCAACAAAAA
	SOC1small-R	CAACAAGAGAGAAGCAGCTTTA
FTM1	FTM1-F	CCCGATGCTATTCGAACATT
	FTM1-R	TCTCTCGTCTGCACGCTCT
FLOR1	FLOR1-F	TCTACGGGAAGATACCACC
	FLOR1-R	AAGGGGAGTTCCACAAAGAC
AP1	qAP1_F	CCTATGGCAATGAGGAGGAA
	qAP1_R	CATCGAACATTTGCCAAAATA
LFY	LFY-RTrw	GGTACGCGAAGAAATCAGGA
	LFY-RTre	ATGACGACAAGCGATGTTCA
Templates for *in situ* probes		
SOC1	SOC1T3-F	ATTAACCCTCACTAAAGGGAATCGAGGAGCTGCAACAGAT
	SOC1T7-R	TAATACGACTCACTATAGGGTTGACCAAACTTCGCTTTCA
FUL	FULT7-R	TAATACGACTCACTATAGGGACGTCTCGACAACGGAGTTC
	FULT3-F	ATTAACCCTCACTAAAGGGAGGGGGAAGATCTTGATTCGT
FTM1	FTM1-F	CCCGATGCTATTCGAACATT
	FTM1-T7-R	TAATACGACTCACTATAGGGTCTCTCGTCTGCACGCT
FTM2	FTM2-F	CAATGTTGATCTGTCCGGTA
	FTM2-T7-R	TAATACGACTCACTATAGGGCGAGAGATAGCGAAATGAGC
FTM3	FTM3-F	TTCAATCCAACGGTGCAG
	FTM3-T7-R	TAATACGACTCACTATAGGGCATTAGCCATAATGGGTTGG
FLOR1	FLOR1-F	TCTACGGGAAGATACCACCA
	FLOR1-T7-R	TAATACGACTCACTATAGGGAAGGGGAGTTCCACAAAGAC
FAF2	FAF2-F	ATTCAGCAGAAGATGCAAGG
	FAF2-T7-R	TAATACGACTCACTATAGGGTCTCCTCAAGAAATCGTACTAAAAA
SPL4	SPL4-F	CCAAAATGGAGGGTAAGAGA
	SPL4-T7-R	TAATACGACTCACTATAGGGCTCCGGTGATACAGTTTTGC
LFY	LFY-F	GGAGCGAGTTACATAAACAAGC
	LFY-T7-R	TAATACGACTCACTATAGGGGCATCCACCACGTCCAGA

Supplemental methods

Analysis of the short-sequence reads for the promiscuous counts

The reads with poly A or T stretches longer than 8 bp or more than one unknown position, N, where discarded. Afterwards, they were trimmed to the first position from the tail with a quality value above 25 with the FASTX toolkit (available at http://hannonlab.cshl.edu/fastx_toolkit/). The trimmed reads were purged from clonal reads, i.e., reads that start with the exact same sequence and could be artifacts from multiple sequencing of the same molecule. The remaining reads were mapped to the TAIR9 genome with bowtie (Langmead et al., 2009), allowing for three mismatches and only uniquely mapping reads were counted. The unmapped reads were remapped with BLAST (Altschul et al., 1990) towards the TAIR9 cDNA sequences in order to extract reads that hit splice junctions.

Many of the gene models in *Arabidopsis thaliana* overlap each other on the genome, e.g., splice variants. While it is not possible to designate the origin of reads mapping within a region present in several gene models, all gene models were clustered on overlap with single linkage. To generate the input data for differential expression analysis, we counted the reads overlapping these clusters, rather than the gene models.

Supplemental references

Langmead, B., Trapnell, C., Pop, M., and Salzberg, S.L. (2009) Ultrafast and memory-efficient alignment of short DNA sequences to the human genome. Genome Biol *10*:R25.

Altschul, S.F., Gish, W., Miller, W., Myers, E.W., and Lipman, D.J. (1990). Basic local alignment search tool. J Mol Biol *215*, 403-410.