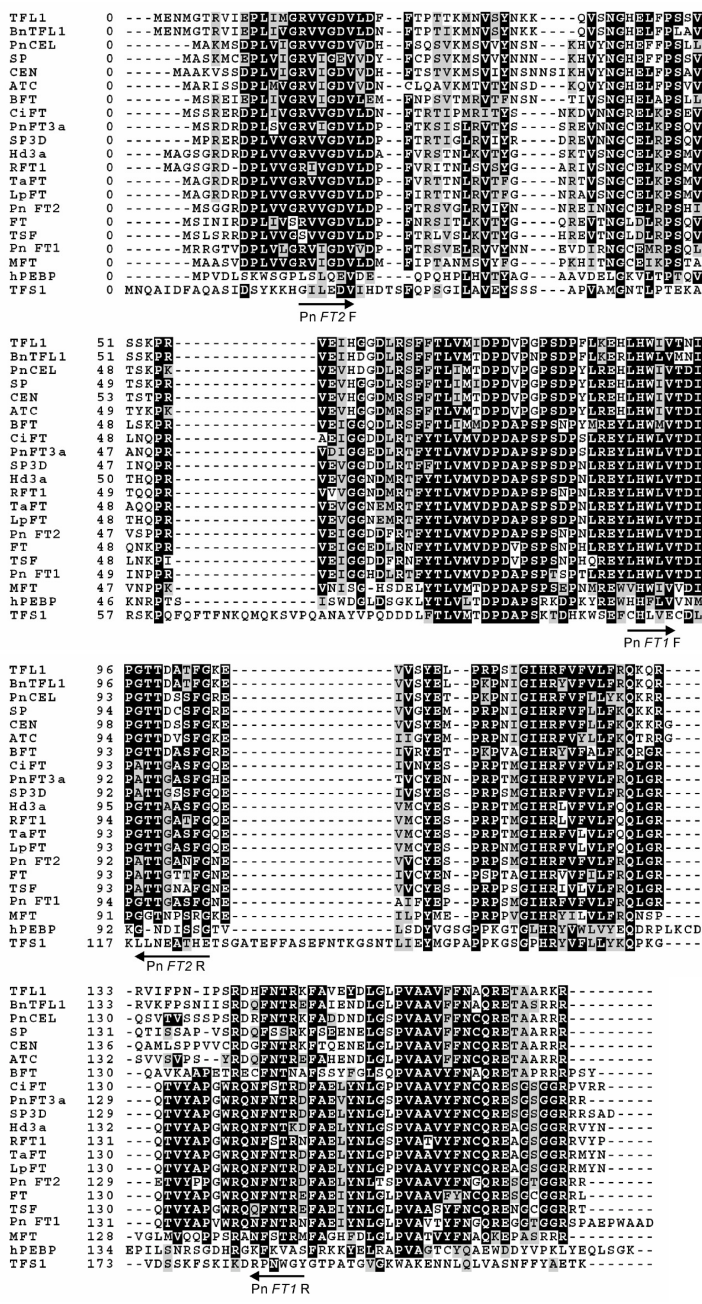
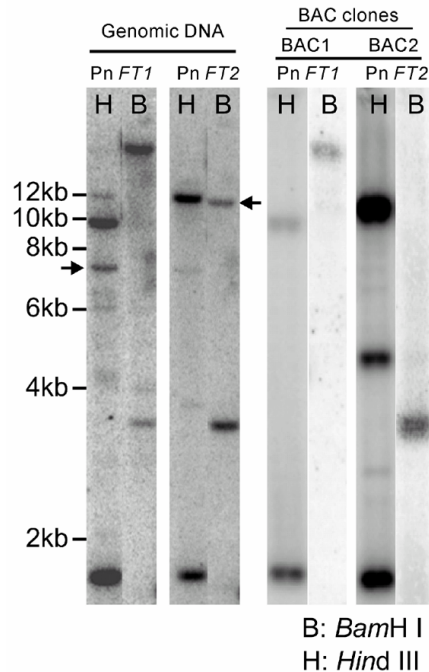


Supplemental figures. Hayama et al., 2007.
 A circadian rhythm set by dusk determines
 the expression of *FT* homologues and the
 short day photoperiodic flowering response
 in *Pharbitis*

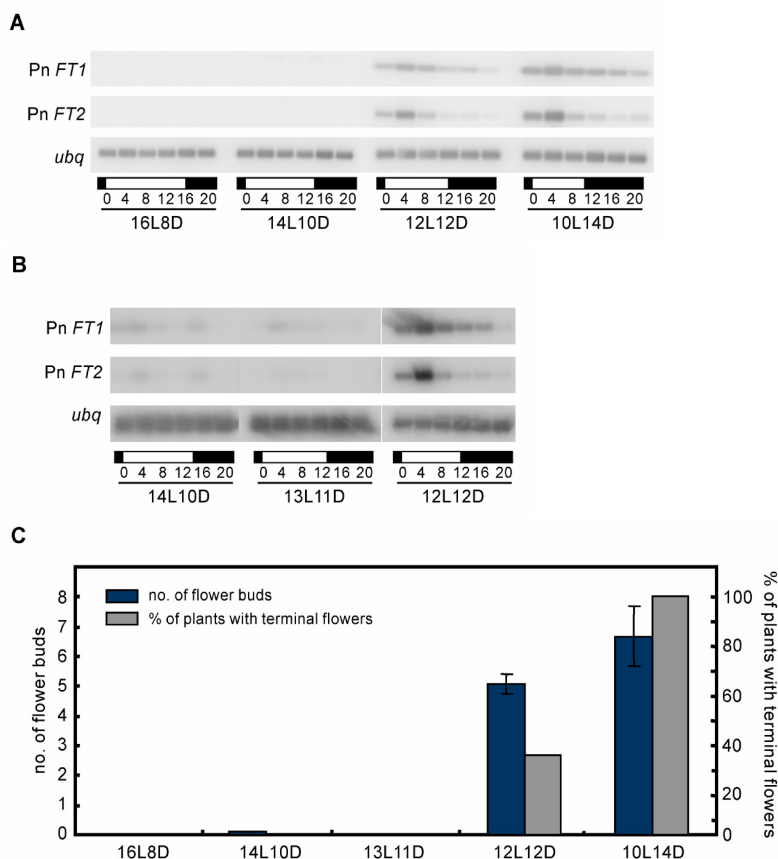


Supplemental Figure 1. Alignment of sequences of FT-like proteins.
 Multiple sequence alignment of the protein sequences used to assemble Figure 1A.
 Positions of the primers used to isolate *Pn FT1* and *Pn FT2* are indicated.



Supplemental Figure 2. Pharbitis genomic DNA and BAC clones of Pharbitis DNA containing Pn *FT1* or Pn *FT2* hybridized with Pn *FT1* or Pn *FT2* probes.

Pharbitis genomic DNA (two left hand panels), a BAC clone containing Pn *FT1* (BAC1) and a BAC clone containing Pn *FT2* (BAC2) were cleaved with *BamH* I (B) or *Hind* III (H). The probes used for hybridization were fragments containing full-length open reading frames of Pn *FT1* or Pn *FT2*, and the probes used in each case are shown above each panel. *BamH* I and *Hind* III fragments characteristic for each gene, as defined by the BAC analysis, are detected in genomic DNA by using Pn *FT1* or Pn *FT2* as a probe. However, additional fragments are detected in the genomic hybridizations that cannot be explained by Pn *FT1* or Pn *FT2*. For example, in the *Hind* III digestion probed with Pn *FT1* 1 (arrow) and in the *BamH* I digestion probed with Pn *FT2* (arrow) one additional fragment that cannot be explained by Pn *FT1* or Pn *FT2* is detected in each case. This analysis indicates that there are likely to be at least two additional *FT*-like genes in Pharbitis.

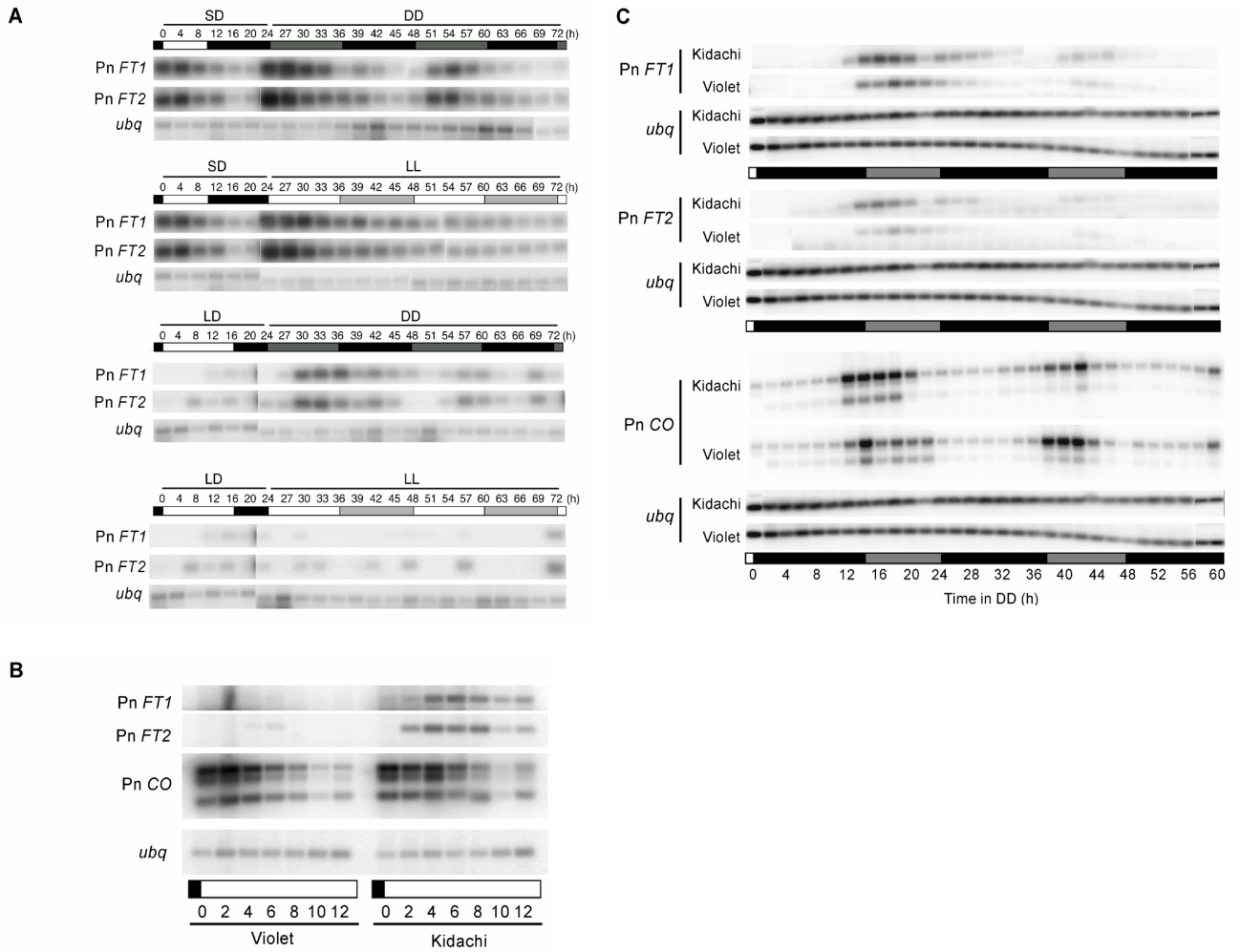


Supplemental Figure 3. Effects of different light/dark regimes on Pn *FT1* and Pn *FT2* expression and on flowering.

A. Pn *FT1* and Pn *FT2* expression under 4 different light/dark regimes. Both genes are expressed under 12L:12D and 10L:14D but not under 16L:8D or 14L:10D. This experiment indicates that a night length between 10 h and 12 h is required to induce expression of Pn *FT1* or Pn *FT2*. RNA was extracted from the cotyledons of plants grown under LDs of 16hL:8hD and then shifted to the conditions shown for three days before harvesting.

B. Pn *FT1* and Pn *FT2* expression under 3 different light/dark regimes. Both genes are expressed under 12L:12D but not under 14L:10D or 13L:11D. This experiment extends the conclusion drawn from panel A, and indicates that a night length between 11 h and 12 h is required to induce expression of Pn *FT1* or Pn *FT2*. RNA was extracted from the cotyledons of plants grown under LDs of 14hL:10hD for 3 days and then shifted to the conditions shown for 4 days before harvesting.

C. Analysis of flowering of plants grown under the 5 different regimes used in panels A and B then shifted to continuous light. Flowering was scored 28 days after transfer to continuous light. Only plants grown under 12L:12D or 10L:14D produce flower buds and terminal flowers. Therefore a precise correlation is observed between those conditions in which Pn *FT1*/Pn *FT2* expression occurs and flowering is observed.



Supplemental Figure 4. Gel blot data used for quantifications shown in Figures.

A. Gel blot data used for Figure 3.

B. Gel blot data used for Figures 5A and B.

C. Gel blot data used for Figure 5 D, E and F.