Mapping of photoperiod-sensitive suppressor of elf3 21
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Abstract:
Arabidopsis thaliana relies on day length or photoperiod to direct the transition from vegetative to reproductive development. The proper response to seasonal cues optimizes growth and reproduction. Many gene products are already known that regulate the photoperiodic control of flowering. Early flowering 3 (elf 3) translates signals from photoreceptors to the circadian oscillator. Floral pathway integrators regulate flowering downstream from the circadian clock. One such integrator, CONSTANS (Co), promotes flowering in long days by activating transcription of Flowering locus T (FT), which results in floral induction. However, the molecular mechanism governing floral induction is not fully understood, and therefore the identification of unknown genes may provide a better understanding. In order to identify novel genes controlling the floral induction pathway, we are characterizing photoperiod-sensitive suppressors of elf3 21. We chose to work on pse21 because it delays flowering of elf3 21 and restores photoperiod sensitivity, but does not cause late flowering on its own. Segregation analysis suggests that there are two pse 21 loci, one on chromosome II and the other on chromosome V. In attempts to understand how pse 21 functions in the floral induction pathway, ongoing studies seek to narrow the location of pse21 on chromosome II and V.

Introduction:

- Many plants optimize their growth and reproduction by sensing and responding to seasonal changes. Day length is one of the most reliable sources that optimize reproductive success in flowering plants.
- Arabidopsis flowering is accelerated by conditions that reliably indicate the passage of winter and the onset of spring and summer (Simpson et al., 2002).
- Arabidopsis thaliana is a useful model for understanding the molecular pathway that regulates flowering in agricultural species.
- A. thaliana contains several known genes that regulate the photoperiodic control of flowering. However, the molecular mechanism governing floral induction is not fully understood and therefore the identification of unknown genes may provide a better understanding.
- In order to find novel genes involved in regulating flowering in Arabidopsis, a screen was done to look for mutations that suppress the elf3 21 flowering time. We are characterizing pse21 to better understand how this mutation alters elf3 1 flowering time in response to day length.
- There are two pse21 loci because we have observed 15:1 segregation from pse21 (Co) x elf3 (Co) progeny to elf3 21 (Co) x elf3 21 (Ws) cross will help us determine the linkage relationship between pse21 DNA and genetic markers of known locations.

Methods:

- Stratification, Planting, and Plant Growth: Seeds were stratified for 2-5 days at 4°C to increase germination rate. For phenotypic analysis, seed was sown on 4" pots filled with most Promix 8X and covered with domes. Plants were grown in short day conditions and remained covered until vegetative leaves began to form. Late flowering plants were transplanted to long day conditions to stimulate flowering and seed set.
- Determination of flowering time: Flowering time was measured by counting the number of rosette leaves produced before stem elongation (bolting).
- DNA Extraction: DNA was extracted as in the CTAB Mini-prep protocol.
- PCR and Gel Electrophoresis: The PCR products were visualized using a 3% agarose gel to assess the genotype at molecular markers of known locations.

Results and Discussion:

- We identified regions containing pse21 on chromosomes II and V using primers of known locations. We measured recombination events between pse21 and ELF3 and I 9602 on chromosome II, and measured recombination events using nga 139, nga 361, ATHCTR1, CAT2, nga139 and LUSSLP02 on chromosome V. On chromosome II, the nga 139 interval overlaps the ELF3 interval suggesting that pse21 is between 27.2 cm and 34.58 cm. On chromosome V, nga 76 and LUSSLP02 argue that pse21 is located between 11.65 cm and 19 cm, respectively. LUSSLP02 and ATHCTR1 argue that pse21 is located between 51 cm and 67.66 cm, respectively (See dashed lines, Figure 5).
- In order to narrow the region containing pse21 on chromosome II we want to use additional markers which include M246, nga 361, AT1H82, and phyB/hy3. Further studies should increase the sample size of mapping population by obtaining different plant families.

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