Online Proceedings

Session 1I

McNair Session - Exploring Science from Cells to Exoplanets

Session Moderator: Janneke Hille Ris Lambers, Biology
MGH 254
12:30 PM to 2:15 PM
* Note: Titles in order of presentation.

Effect of Daily Temperature Changes in *FLOWERING LOCUS T* Expression Profiles in Nature
Dianne Laboy, Junior, Pre-Sciences
Levinson Emerging Scholar, McNair Scholar
Mentor: Takato Imaizumi, Biology
Mentor: Akane Kubota, biology

Climate change is forecasted to have an impact in agricultural productivity. To ensure security of crop yields, we must develop a better understanding of flowering mechanisms in plants. Research on flowering mechanism has been performed mostly under simplified lab growing conditions. However, in the natural environment, plants experience dynamic changes in abiotic factors like temperature throughout the day. To better understand how flowering is regulated under natural conditions, we focused on the expression profiles of *FLOWERING LOCUS T* (*FT*), which correlates with the flowering time in plants. *FT* expression is known to peak in the evening under long day lab conditions (16 hours of light, 8 hours of darkness, 22^{o}C constant). However, under natural long day conditions (Summer Solstice in Seattle, 16 hours of light, 8 hours of darkness, average highest temperature 21^{o}C) we have observed an additional morning peak. We have been able to recreate the observed expression of *FT* in the lab by modulating temperature oscillation and light quality of long day lab conditions. In this project, we first asked if the observed *FT* pattern is widely conserved in different wild type accessions in *Arabidopsis thaliana*. We grew different accessions under re-created lab growth conditions and analyzed *FT* expression using quantitative PCR. So far we have seen similar *FT* expression patterns among some accessions, suggesting that morning peak of *FT* is conserved among these lines. Second, we studied which timing of the day temperature acts as a cue to affect morning *FT* expression. We grew plants under various temperature conditions and compared results to the observed *FT* expression in nature. The results from this project will contribute to the development of new modified lab growth conditions that represent natural conditions and give a better understanding of the mechanism that controls *FT* regulation in nature.

Poster Session 2

Commons East, Easel 84
1:00 PM to 2:30 PM

Regulatory Mechanism of *FLOWERING LOCUS T* Mediated by *PHYTOCHROME A* under Natural Conditions
Jessica Imel, Senior, Biochemistry, Biology (Physiology)
Mentor: Takato Imaizumi, Biology
Mentor: Akane Kubota, biology

Plants respond to abiotic factors in natural settings in order to regulate physical responses at the most appropriate timing. The timing of flowering affects seed and/or fruit production in many agriculturally relevant plants, so understanding its mechanism is essential for addressing agricultural needs as climate changes. To address this aim, we have used *Arabidopsis thaliana* as a model plant to understand the molecular mechanism of seasonal flowering. It has been shown that the expression of *FLOWERING LOCUS T* (*FT*), which encodes florigen, peaks in the evening under long-day lab conditions (16 hours light, 8 hours dark, 22C) and promotes flowering in a day-length specific manner. However, our lab found that *FT* also peaks in the morning under outside long-day conditions over the summer solstice in Seattle (16 hours light, 8 hours dark, highest average temperature 21C). This double-peak of *FT* can be re-created by changing the light quality and temperature settings of long-day lab conditions. In addition, we found that a mutant of *PHYTOCHROME A* (*PHYA*), which encodes a red/far-red light photoreceptor in plants, lacks *FT* expression in a morning-specific manner, suggesting that *PHYA* is involved in regulating morning expression of *FT*. In order to understand the molecular mechanism of *PHYA*-mediated *FT* regulation, we are in the process of identifying transcription factors and other molecules that work with PHYA to induce *FT* expression. We have successfully cloned various transcription factors that are reported to act in the flowering regulatory pathway and are in the process of testing protein-protein interaction against PHYA by

yeast-two-hybrid methods. Identifying the components that act in the *PHYA-FT* pathway will allow us to further understand how light affects flowering regulation.

SESSION 2P

ENVIRONMENTAL RESPONSE OF PLANTS AND THE UNDERLYING MECHANISMS

Session Moderator: Takato Imaizumi, Biology
JHN 022
3:30 PM to 5:15 PM
* Note: Titles in order of presentation.

Kinetics of FKF1 LOV Chemistry Determine its Function in Photoperiodic Flowering in *Arabidopsis*
Simone Lee (Simone) Kemp, Senior, Biology (Molecular, Cellular & Developmental)
Mentor: Takato Imaizumi, Biology
Mentor: Jaesung Shim, Department of Biology

FLAVIN-BINDING KELCH REPEAT F-BOX 1 (FKF1) is a blue light photoreceptor which induces photoperiodic flowering in *Arabidopsis*. Depending on blue light, FKF1, along with GIGANTEA, forms a complex to induce flowering by degrading a transcriptional repressor, CYCLING DOF FACTOR 1 (CDF1). FKF1 stays in light status for a long time, allowing plants to continue induce flowering. ZEITLUPE (ZTL), a close homolog of FKF1, is a circadian clock regulator with a faster photocycle LOV domain. A photocycle is the sequence of structural changes that takes place in molecules when exposed to light. ZTL has high affinity with GI in presence of light, but also degrades its targets quickly in the dark. The faster ZTL photocycle allows it to act as a sensor to the night periods in circadian oscillation phases. Therefore, we hypothesize that slower photocycles of FKF1 is necessary for FKF1 to keep light information and induce photoperiodic flowering. To test the effect of photocycle on the FKF1 LOV domain in photoperiodic flowering, we generated mutations on the FKF1 LOV domain based upon sequence information of the ZTL LOV domain, to speed up the photocycle kinetics. Quantitative real time PCR (qPCR) was used to select stable, transgenic lines expressing comparable levels of mutated FKF1. Then we analyzed samples for expressions of downstream genes *CONSTANS, FLOWERING LOCUS T*, along with flowering times to measure FKF1 functionality. As a control, wild type plants were used to provide regular flowering time data. We gathered samples to establish lines to test the effect of each mutation on FKF1 function. We expect to find that the mutations' faster conversion to dark status decreases its affinity to GIGANTEA and CDF1 degradation, delaying the flowering time. This suggests the slow conversion of FKF1 LOV domain to dark status is necessary to induce photoperiodic flowering in *Arabidopsis*.

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ENVIRONMENTAL RESPONSE OF PLANTS AND THE UNDERLYING MECHANISMS

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CDF1 Utilizes TPL to Repress Photoperiodic Flowering in Arabidopsis
Evan Groover, Senior, Biology (Molecular, Cellular & Developmental)
UW Honors Program
Mentor: Greg Golembeski, Biology
Mentor: Takato Imaizumi, Biology

Properly timed seasonal flowering in plants is necessary for reproductive viability in natural and agricultural settings. We use *Arabidopsis thaliana* as a model organism to study the molecular mechanisms that underlie the developmental transition to flowering, a process that is mediated by both internally propagated circadian cycles as well as information about day length (photoperiod) detected via photoreception. Transcriptional regulation of the *FT (FLOWERING LOCUS T)* gene is the primary mechanism of controlling the flowering response, and flowering occurs when requisite levels of FT proteins are transported from the leaves to apical stem cell tissue. *FT* expression is initiated by CO (CONSTANS), a transcription factor whose own expression is tightly regulated by molecular outputs of the endogenous circadian clock. One such output, *CDF1 (CYCLING DOF FACTOR 1)* is responsible for repression of *CO* and *FT* expression. We established that CDF1 forms a complex with the TPL (TOPLESS) protein, an interaction that is dependent on a conserved peptide sequence at the N-terminal of CDF1. We generated phloem companion tissue-specific mutants of *TPL* in a variety of flowering time genetic backgrounds and found that mutants of *TPL* consistently flowered earlier than nonmutants, similar to the phenotype witnessed in *cdf* mutants. *tpl* mutation had a similar effect on *CO* and *FT* expression across 24-hour timecourse to *cdf-1,2,3,5* mutants in both long and short day conditions. We have shown that TPL protein associates with *CO* and *FT* promoters during the mornings of long days, and that this recruitment of TPL to the promoters is enhanced by the addition of excess CDF1 protein. We demonstrate that TPL protein is required for the repressive activity of CDF1 and are working to better understand the complex role it plays

in regulating flowering time in *Arabidopsis*.