Online Proceedings

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.

Changes in Caloric Value of Algae Digested by the Sea Urchin *Mesocentrotus franciscanus* and Implications for Benthic Communities

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Mary Gates Scholar, UW Honors Program

Mentor: Megan Dethier, Biology

Algal subsidies are extremely important to the success of the deep benthos where there is little to no primary productivity. Little research has been done on the nutritional value of detritus, such as pieces of kelp, that sink into deep habitats. Sea urchin feces, as a form of detritus, may provide an energetic link from shallow-water algae to these deep benthic communities. Urchins are known to have an inefficient digestive system which creates the potential for feces to be of high food value. In this study, relative caloric contents of algae and of feces of the red sea urchin (Mesocentrotus franciscanus) were analyzed by chemical assay. Algal biomass consumed and feces egested by urchins were quantified, as were the caloric contents of fresh and aged urchin feces fed a diet of bull kelp (Nereocystis luetkeana). In all cases, the caloric content of algal material increased after being consumed by urchins, and the longer the urchin feces aged, the higher the caloric value became. It is likely that microbiota inside urchin guts are driving these counterintuitive results. The creation of calorie-rich feces could add to the importance of urchins as a link to benthic communities that rely heavily on detritus for their success.

Quantifying Leaf Phenotype of AFB Mutants in *Arabidopsis thaliana*

Mollye Lucile Zahler, Junior, Biology (General)

Mary Gates Scholar

Mentor: Jennifer Nemhauser, Biology

Mentor: Clay Wright, Biology

The plant hormone auxin controls nearly every aspect of plant growth and development. Proteins involved in the perception of auxin are closely studied in an effort to elucidate drivers of plant morphology. Potential applications of such research lie in engineering of plant architecture and ultimately maximizing crop yield. Auxin triggers binding between two proteins: auxin receptors (Auxin-signaling F-Boxes or AFBs) and transcriptional repressors (Aux/IAAs). This interaction leads to the ligation of ubiquitin to Aux/IAAs, triggering their degradation, and allowing transcription of auxin-induced genes. In the model plant *Arabidopsis thaliana*, the AFB family has six members (TIR1 and AFB1- AFB5). While the AFBs are highly similar in sequence to one another, individual AFBs may play specialized roles. In our preliminary studies, we have found that a reduction in AFB2 activity in the *afb2-3* mutant produces a leaf phenotype not observed in *TIR1* mutants. To confirm that *AFB2* is responsible for this phenotype, we are engineering transgenic *afb2* mutants that express a wild-type copy of AFB2. We expect that leaves of these transgenic plants will resemble those of wild-type. We are also analyzing the leaf phenotypes of other AFB mutants. To test whether small differences in AFB2 function can change leaf phenotype, we are currently transforming natural hypomorphic and hypermorphic AFB2 variants into *afb2-3* mutants. By quantifying leaf morphology of these transgenic plants, we plan to map the relationship between *AFB2* function and leaf phenotype.

CDF1 Utilizes TPL to Repress Photoperiodic Flowering in Arabidopsis

Evan David 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*.

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*.