

Paper for Tuesday October 22, 2012

Kumar SV, Wigge PA. 2010. H2A.Z-containing nucleosomes mediate the thermosensory response in Arabidopsis. Cell. Jan 8;140(1):136-47.

Questions for Thought

Before you start reading this paper, think of possible molecular mechanisms to sense changes in ambient temperature. How do humans do it? What do we sense? What is a problem that organisms more similar to plants face?

As before, **write down questions** you have about the logic or rationale for each experiment, the method employed, and the conclusions drawn. Come up with at least three questions. Turn in your questions as homework at the beginning of class. During class we will discuss your questions along with the QfT below. We will emphasize the questions in bold; the other questions are meant to help you think about each issue.

- 1) Ambient temperature is critical for plant development and yet little is known about how plants perceive temperature. The authors decide on a forward genetic screen. **Why?** What other approaches could they have taken?
- 2) The authors develop a reporter system for their screen based on a particular HSP70. **Why are the comparisons shown in Fig. 1B and 1C critical for the success of their screen?** What are the advantages of using luciferase as a reporter compared to more conventional reporters such as GUS and LacZ?
3. The authors use transcript-based cloning to identify the mutated gene. **What are potential pitfalls of this method?** How do the authors establish that *entr1* and *entr2* are alleles of the same gene?
4. **What genetic experiments could the authors have conducted to further explore the role of ARP6, PIF4, and various H2A.Z in ambient temperature response?** What results would you predict and why?
- 5) H2A.Z is evicted in wild-type plants in response to higher temperature. **How would you start to address how this eviction occurs and how it is regulated?** Genetically? Biochemically?
- 6.) Researchers have collected genetically divergent *A. thaliana* strains from around the globe. **How would you make use of this resource to further investigate thermosensing and in particular the mechanism(s) of H2A.Z eviction?**
7. A simple explanation for temperature-sensitive eviction of H2A.Z would be that this histone is thermolabile. **Why is this unlikely?** Compare with the cold-regulated process of inducing flowering that is briefly described in the introduction/discussion.