GS351 Fall 2006 Preparatory Questions for Tuesday October 17th

Assigned readings:

Carey, M. (1998) The Enhanceosome and Transcriptional Synergy. Cell 92: 5-8.

Kim T and T Maniatis (1997) The Mechanism of Transcriptional Synergy of an In Vitro Assembled Interferon-β Enhanceosome. Molecular Cell 1:119-129.

Optional background readings:

Hahn S (2004) Structure and mechanism of the RNA polymerase II transcription machinery. Nature Structural and Molecular Biology 5:394-403. (Review)

Ptashne M and A Gann (1997) Transcriptional activation by recruitment. Nature 386:569-577. (Review)

1. What is synergy? How is it assayed? What are its components?

2. Describe how Kim and Maniatis developed an in vitro transcription system for assaying the enhanceosome. What results in Fig 1 demonstrate synergy? Why?

3. What is HMG I(Y)? How does it differ from ATF2, c-JUN, IRF1, p50 and p65?

4. How was it shown (Fig 2) that HMG I(Y) is required for transcriptional synergy?

5. How was it shown (Fig 3A) that a specific interaction occurs between HMG I(Y) and ATF2?

6. How was it shown (Fig 3B) that HMG I(Y) binding to DNA is also important?

7. What experiments were done (Fig 3C) to show that the relative orientation of each of the PRDs is important?

8. How was the contribution of increased stability to the mechanism of synergy of the enhanceosome shown?

9. What is cooperativity and how is it measured? How was cooperativity shown (Fig 5) to be important for the assembly of the IFN β enhanceosome?

10. What is sarkosyl and how was it used (Fig 6 and Fig 7) to investigate the mechanism of enhanceosome function?

11. Discuss the mechanism proposed for function of the INF β enhanceosome. What other functions might the enhanceosome have?