

Paper for Tuesday 13 January 2009

Hereford, L. M. and Hartwell, L. H. 1974. Sequential gene function in the initiation of *Saccharomyces cerevisiae* DNA synthesis. *J. Mol. Biol.* **84**: 445-461.

Questions for Thought

As you read this paper, **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) Summarize the state of knowledge at that time. What is the pathway they are studying? What are the possible choices a yeast cell might make at this stage of the cell cycle? What was the purpose of these experiments? **Why did they choose these genes to study?** What was known about these genes relative to S phase? relative to alpha factor? What are their phenotypes? What are the two general approaches that they take to achieve their purpose and how do they differ? How did the goal of these experiments affect their choice of genes?
- 2) Table 1: **Explain the logic of the reciprocal shift experiments.** What are essential aspects of these experiments? What assumptions are they making about the pathways involved in this process? What are “restrictive conditions”? What is meant by “terminal phenotype”?
- 3) Figure 1: Explain the result with *cdc4*. What exactly are they assaying? What if their only assay was cell division? **How do they know how long to leave the cells at 36°C?** What if the blocks to cell cycle advancement are slowly imposed?
Note: we will focus on Figure 1. Figures 2 and 3 give similar results.
- 4) Table 2: **Explain the logic of the double mutant studies.** What is an essential aspect of the mutant phenotypes for this experiment to work? **What if they don't have null alleles?** Why does the *cdc28* allele give such a variable phenotype?
- 5) Table 3: Summarize their results. How do they put *cdc4* before *cdc7*? **What evidence corroborates their inferred gene order?**
- 6) **Summarize the logic of these methods and indicate the restrictions or caveats to each approach.**