

BE/Bi 101: Order-of-Magnitude Biology
Homework 3
Due date: Friday, April 22, 2016

“Almost all aspects of life are engineered at the molecular level, and without understanding molecules we can only have a very sketchy understanding of life itself.”

—Francis Crick in *What Mad Pursuit*

1. Protein to mRNA ratio.

One of the intriguing quantities that characterizes the central dogma is the protein to mRNA ratio. A simple model of protein synthesis is of the form

$$\frac{dp}{dt} = \beta m - \alpha p, \tag{1}$$

where β is the rate of protein synthesis per mRNA molecule and α is the protein degradation/dilution rate. The quantities m and p represent the mRNA and protein concentrations. Work out the steady state value of the protein to mRNA ratio, p/m , based on the necessary counts and growth rates we discussed in class. Separately, estimate the parameters β and α and use them to evaluate the steady state protein to mRNA ratio approximately. Do your two estimates agree?

2. How many polymerases?.

This past Wednesday, I (JB) was lecturing in BE 150/Bi 250b. We were talking about strong versus weak promoters, and we had to make an impromptu estimate of the number of polymerases in a bacterial cell. I started doing some street-fighting estimations, but I couldn't get an estimate faster than Prof. Elowitz could look it up on Bionumbers. Nonetheless, I think it is a fun and instructive estimate to make. Street-fight your way to that estimate.

3. RNA polymerase and ribosomes.

This is essentially problem 3.4 from Physical Biology of the Cell, 2nd Ed., by Phillips, Kondev, Theriot, and Garcia.

- a) If RNA polymerase subunits β and β' together constitute approximately 0.5% of the total mass of protein in an *E. coli* cell, how many RNA polymerase molecules are there per cell? The subunits have a mass of 150 kDa each. This part of the problem was adapted from Problem 4.1 of Schleif, *Genetics and Molecular Biology*, 1993.
- b) Rifampin is an antibiotic used to treat Mycobacterium infections such as tuberculosis. It inhibits the initiation of transcription, but not the elongation of RNA transcripts. The time evolution of an *E. coli* ribosomal RNA (rRNA) operon after addition of rifampin is shown in Fig. 1. An operon is a collection of genes transcribed as a single unit. Use the

figure to estimate the rate of transcript elongation. Use the beginning of the “Christmas-tree” morphology on the left of Fig. 1(A) as the starting point for transcription.

- c) Using the calculated elongation rate, estimate the frequency of initiation off of the rRNA operon. These genes are among the most transcribed in *E. coli*.
- d) As we saw in the class, a typical *E. coli* cell with a division time of 3000 seconds contains roughly 20,000 ribosomes. Assuming there is no ribosome degradation, how many RNA polymerase molecules must be synthesizing rRNA at any instant? What percentage of the RNA polymerase molecules in *E. coli* are involved in transcribing rRNA genes?

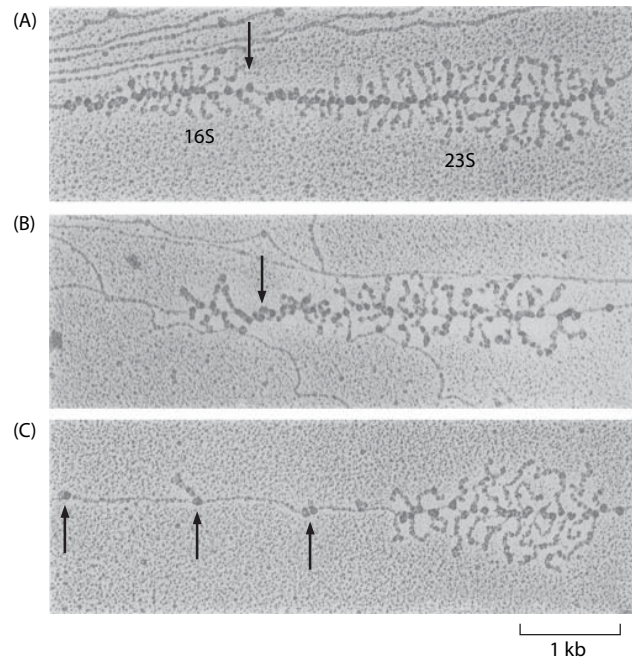


Figure 1: Effect of rifampin on transcription initiation. Electron micrographs of *E. coli* rRNA operons: (A) before adding rifampin, (B) 40 seconds after addition of rifampin, and (C) 70 seconds after exposure. No new transcripts have been initiated, but those already initiated are carrying on elongation. In parts (A) and (B) the arrow signifies the site where RNaseIII cleaves the nascent RNA molecule producing 16S and 23S ribosomal subunits. RNA polymerase molecules that have not been affected by the antibiotic are marked by the arrows in part (C). This is Fig. 3.29 of *Physical Biology of the Cell, 2nd Ed.*, by Phillips, Kondev, Theriot, and Garcia, which was adapted from L. S. Gotta et al., *J. Bacteriol.* 20:6647, 1991.)