

BE/APh161: Physical Biology of the Cell

Homework 1

Due Date: Wednesday, January 11, 2017

“A physics that has no place for life is as impoverished as would be a biology not informed by chemistry. The study of life as a natural phenomenon, a fundamental feature of the universe, must not be allowed to slip into the black hole of departmental tribalism.” - Franklin Harold, *The Way of the Cell*

When doing street fighting estimates, the goal is to do simple arithmetic of the kind that all numbers are 1, few or 10. $\text{few} \times \text{few} = 10$, etc. Please do not provide estimates with multiple “significant” digits that are meaningless. Be thoughtful about what you know and what you don’t know. You may use the Bionumbers website (<http://bionumbers.hms.harvard.edu/>) to find key numbers (examples are masses of amino acids (BNID 104877) and nucleotides (BNID 103828), the speed of the ribosome (BNID 100059), etc.), but please provide a citation to the Bionumber of interest as shown above. However, for many of these problems the essence of things is to do simple estimates, not to look quantities up.

1. To build a cell.

Do problems 2.5 (ingredients in minimal media) and 3.7 (sugar budget of a cell) from PBOC2. Together, these two problems are intended to get you thinking about the wondrous process whereby a clear liquid with simple chemical ingredients is converted into biomass as shown in Figure 1. Amazing! In addition to working out the two problems given above, also work out an estimate for the volume of the headspace you see in Figure 1 which has oxygen available for cell growth. Specifically, if 6 O_2 molecules are consumed for every sugar, make a simple estimate of the required volume of headspace needed to sustain cell growth. Note that our estimate about O_2 usage is crude and sloppy. To really do this carefully, we need to acknowledge the use of glucose both in providing building materials (i.e. carbon skeletons) as well as the energy needed to synthesize a cell. The estimate we have done here is intended to give an impression of the magnitudes, and specifically to get a sense of the aeration requirements when we do a liquid culture growth procedure.



Figure 1: Growth of *E. coli* in rich media. The tube on the left shows roughly 5 mL of growth media just after inoculation. The tube on the right shows such media after saturation due to exponential cell growth and division.

2. Proteomic data on bacteria in different growth conditions.

Read the paper by Schmidt and Heinemann and co-workers in which they use mass spectrometry to take the census of *E. coli* under a variety of different growth conditions. The outcome of this work was a census of the number of copies of roughly half of the proteins in this important bacterium.

(a) Using the data in the spreadsheet available with this homework, examine the numbers for the subunits of ATP-synthase. Write a short paragraph describing what ATP synthase is and what it does. Then, make an estimate of the number of ATPs it takes to make a new cell. In light of the number of ATP synthases counted by Heinemann and his group, are there enough to make all the ATPs needed to build a cell?

(b) Comment on the units on the y-axis of figure 2b of the Schmidt *et al.*

paper. Specifically, justify those units in terms of what you know about the total number of proteins and the mass per protein. Do you think that the measurements pass the street fighters sanity check? Explain your conclusions.

3. RNA Polymerase and Rate of Transcription.

One of the ways in which we are trying to cultivate a “feeling for the organism” is by exploring the processes of the central dogma. Specifically, I want you to have a sense of the number of copies of the key molecular players in the central dogma as well as the rates at which they operate. Further, I argue that it is critical you have a sense of *how* we know these numbers. To that end, to get a feeling for transcription, do problem 3.4 of PBoC2.

4. Street fighting your way to the ribosome density.

One of the most important molecular assemblies in the cell is the ribosome. The number of ribosomes per cell dictates how fast cells can grow with *E. coli* growing with a division time of 24 minutes having 72,000 ribosomes per cell and slow growing *E. coli* with a division time of 100 minutes having a factor of ten fewer ribosomes with a count of ≈ 6800 ribosomes. In this problem, we will use our street fighting skills to explore the ribosomal density in another organism as shown in Figure 2, and then see how well our results from the electron microscopy study square with the numbers quoted above. By examining the figure, make an estimate of the number of ribosomes per μm^3 and compare that result to the numbers quoted above.

5. RNA vs Protein

Using the kind of estimates we have talked about in class, give a simple characterization of the relative sizes of mRNAs and the proteins they code for. Specifically, first comment on the mean mass of amino acids and nucleotides as well as their typical physical sizes. Use both of these metrics as a way to provide a rough sense of the relative sizes both in mass and physical dimensions of proteins and the mRNAs that code for them.

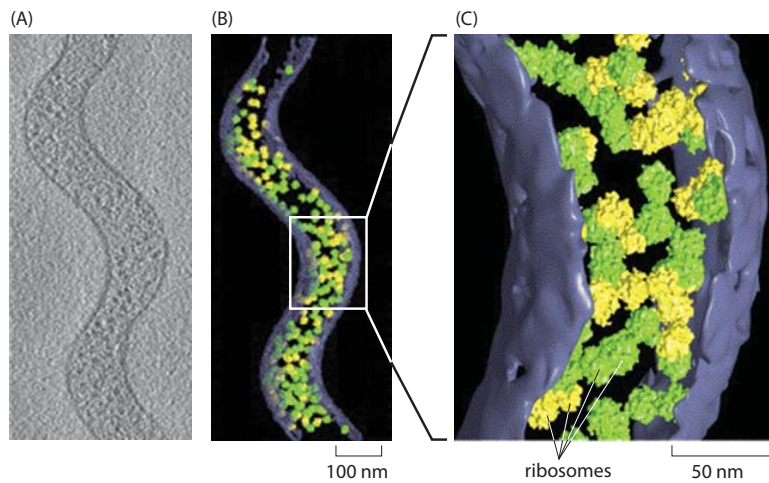


Figure 2: Cryo EM study of a bacterial cell. These images are of the tiny bacterium, *Spiroplasma melliferum*. Using algorithms for pattern recognition and classification, components of the cell such as ribosomes were localized and counted. (A) Single cryo-electron microscopy image. (B) 3D reconstruction showing the ribosomes that were identified. Ribosomes labeled in green were identified with high fidelity while those labeled in yellow were identified with intermediate fidelity. (C) Close up view that you should use to make your count. Adapted from JO Ortiz *et al.*, J. Struct. Biol. 156, 334-341 (2006).