BE/Bi 101: Order-of-Magnitude Biology
Homework 5
Due date: Friday, May 13, 2016

"Probability theory is nothing but common sense reduced to calculation."

—Pierre-Simon Laplace

1. Dynamics of Earth and evolution.
One of the reasons that evolution is hard to think about is because of the vast times involved in evolutionary processes. One of the more interesting things we have to remember is the interplay between the dynamics of the earth and the dynamics of the living organisms on earth. For example, think about the consequences of the closing off of the Isthmus of Panama for evolution. Similarly, the collision of India with the Asian continent brought an end to the Tethys Sea. In this problem, we will think about such geological processes as they bear on evolution.

We begin by thinking about how fast the Galapagos Islands are moving. You should look up the relevant geographical information about these islands such as their positions and elevation. Note that these islands, like the Hawaiian Islands, are being produced by a "hotspot" that is near the current islands of Isabella and Fernandina. The islands then move in a southeasterly direction towards the coast of South America. Given that the island of Espanola is roughly 3.5×10^6 years old, make an estimate for the mean rate at which these islands are moving to the southeast per year. Give your answer in cm/year. Also, notice that as the islands age, their height is reduced with the new islands of Fernandina and Isabella with volcanoes over 1500 m in height while Espanola has a height of only roughly 200 m. Espanola is the easternmost island in the Galapagos. Yet, the divergence time for the different species of finch on the islands appears to be more than 3.5×10^6 years. How can you make sense of this? Assuming that the speed you found for the Galapagos is typical for island chains, make an estimate of the age of the island Kauai using the same kind of logic. What factors might complicate this comparison, and how would they change this estimate (qualitatively)?

2. Most abundant protein on Earth.
In class we mentioned that there are often conflicting views on the most abundant proteins on Earth. Currently, the best guesses that we have been able to come up with are histones, collagen and RuBisCO. In this problem, you will try to come up with estimates for each of these protein counts to draw your own conclusions about protein abundance.

a) We'll start with estimates of histones. Recall that histones are the proteins that make up nucleosomes, the architectural units that are responsible for wrapping up the DNA in eukaryotic nuclei. To be concrete, how long is the human genome in meters? If 147 bp are wrapped around each histone octamer to make up a nucleosome, and the spacing between nucleosomes is between 25 and 100 bp, how many nucleosomes (and histone proteins) are

there for each of our cells? How many cells do we have and hence, how many histones are there per human? Given that all eukaryotes use these architectural proteins, you will have to figure out a strategy to estimate the total number of eukaryotes and thereby, the amount of histone protein. Estimate the number of copies and total mass of histone proteins on Earth.

b) Collagen is one of the key protein components of the extracellular matrix, the set of molecules that are secreted by cells and are known to provide mechanical support. Estimate the number of copies of collagen proteins on Earth, or alternatively, the total mass of collagen. Write a few sentences about what role collagen plays in animals and then give your rationale for your estimate about the total amount of collagen protein based on the fraction of animals that is extracellular matrix and how collagen fits into that picture.

c) We are going to do the estimate of the amount of RuBisCO using a method may seem relatively tortuous, but will give us a chance to think about the relationship between photosynthesis and the atmosphere. We begin by trying to figure out how much CO_2 is used in photosynthesis over the course of a year by looking at the Keeling curve (see Figure 1). Begin by estimating the mass of the atmosphere using

$$M_{\text{atmosphere}} g = p_{\text{atmosphere}} A_{\text{earth}}. \tag{1}$$

Explain why this equation is an appropriate way to estimate the atmospheric mass. Next, using the average mass of the molecules in the atmosphere, figure out how many molecules there are in the atmosphere and then using the Keeling curve, figure out how many CO_2 molecules there are. To determine the number of CO_2 molecules fixed each year in the process of photosynthesis, we make a crude approximation based on the annual variations in the Keeling curve, which reflect an asymmetry in the photosynthetic output of the northern and southern hemispheres. In particular, we note that the variability in atmospheric CO_2 is roughly ten parts per million and we make the guess that this variability is comparable to the scale of net carbon fixation itself. Work out how much total CO_2 is fixed on the basis of the Keeling curve, both in number of molecules and in total mass of carbon. We can now make naive estimates about the number of RuBisCOs by using the rough figure that each such RuBisCO on average fixes one carbon per second. Given the total amount of CO_2 fixed per year (based on the Keeling curve in the way described above) and the rate at which the enzyme works, how many copies of RuBisCO do you estimate there are on Earth?

3. Using probability to figure out how molecular motors walk.

Kinesin is a motor protein that "walks" in a directed fashion along microtubules. A little more than a decade ago, it was unknown whether kinesin walked along microtubules hand-over-hand or like an inchworm (See Fig. 2).

To settle this question, Yildiz and coworkers performed an elegant experiment. They tagged one of the "heads" of kinesin with a fluorescent dye. They then used total internal reflection

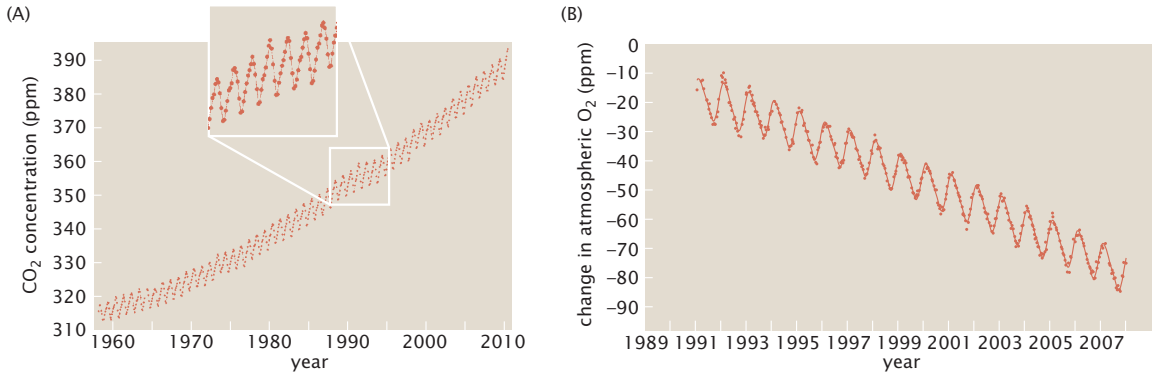

Figure 1: The Keeling curve showing atmospheric (A) CO_2 concentration and (B) O_2 concentration in parts per million. Adapted from *Physical Biology of the Cell* by Phillips, Kondev, Theriot and Garcia.

fluorescence microscopy (TIRF) to monitor the movement of the fluorescent dye over time.[1] By performing a curve fit of the fluorescent signal in their images with a Gaussian (which approximates the point spread function of the fluorophore), they can pinpoint the position of the fluorescent tag to an accuracy close to one nanometer. Such a curve fit is shown in Fig. 3.

The dynamics of the motor is revealed in traces like those shown in Fig. 4. From these traces, they can compute the step length (the height of the vertical segments of the red lines) and the dwell time of the labeled head between movements (the length of the horizontal segments of the red lines).

a) Based on Fig. 5A, does kinesin more likely walk hand-over-hand or like an inchworm? Justify your answer.

b) In the schematic of hand-over-hand motion shown in Fig. 2, each step consists of detachment of the back foot and its subsequent reattachment in the front position. Which do you think happens more rapidly, detachment or attachment of the head? Explain your reasoning.

c) To provide further evidence in favor of the hand-over-hand model to describe kinesin walking, we can investigate the dwell time of the fluorophore between displacements. The distribution of the dwell times of the fluorophore is shown in Fig. 5B. Based on this histogram, does kinesin more likely walk hand-over-hand or like an inchworm? Use the stories of probability distributions to justify your answer.

After completing this problem, it is well worth reading the paper, which you can download here.

[1] You can see a movie of one of their fluorescent dyes, corresponding to the top trace of Fig. 4, here. The interpixel spacing is 86.6 nm. The movie lasts 20 seconds in total, with a frame rate of 3 frames per second.

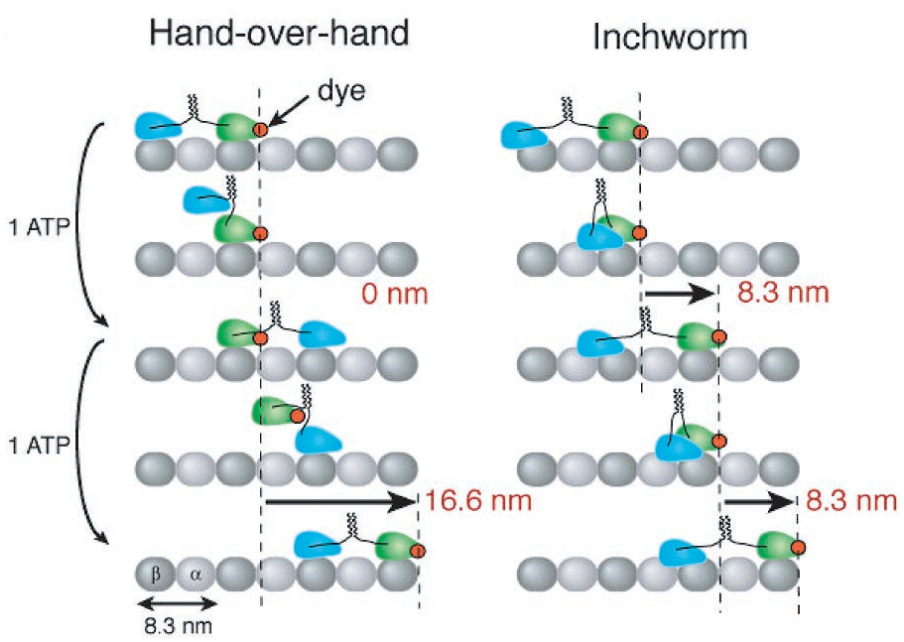

Figure 2: Schematic of hand-over-hand and inchworm mechanisms of kinesin walking on microtubules. Kinesin heads bind to the β subunit of the tubulin dimers that comprise a microtubule. Thus, the microtubule geometry dictates kinesin step size that is an integer multiple of the spacing between β subunits, 8.3 nm. Adapted from Yildiz, et al., *Science*, **303**, 676–678, 2004.

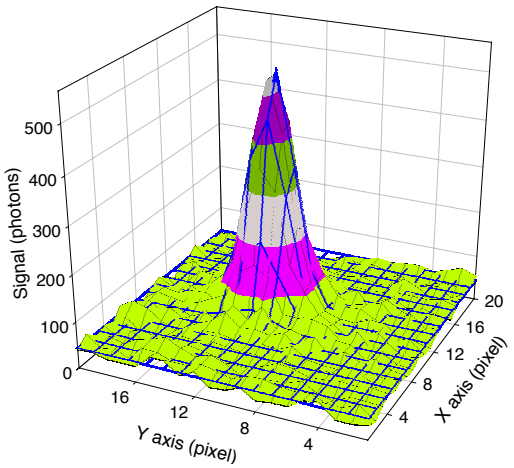

Figure 3: Plot of photon counts (colored surface) with Gaussian fit (blue mesh). The position of the maximum of the fitted Gaussian is used to pinpoint the position of the fluorophore. Figure taken from Yildiz, et al., *Science*, **303**, 676–678, 2004.

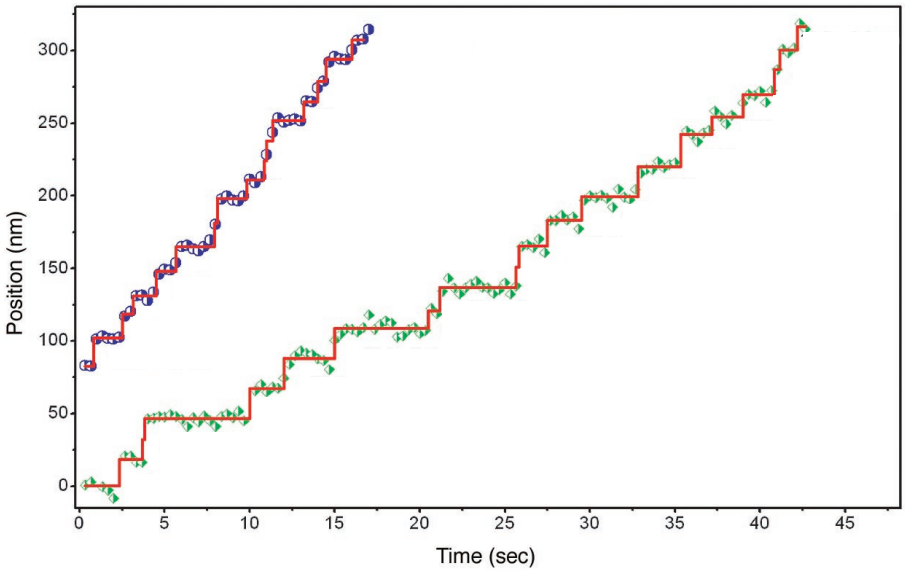

Figure 4: Sample traces of the position of the fluorophore versus time. Symbols are measurements; red lines are the results of data analysis. The steps (vertical red lines) are typically faster than the 0.5 s frame rate of the experiment, so they appear instantaneous. These traces are for homodimer kinesins (with only one of the feet fluorescently labeled). Adapted from Yildiz, et al., *Science*, **303**, 676–678, 2004.

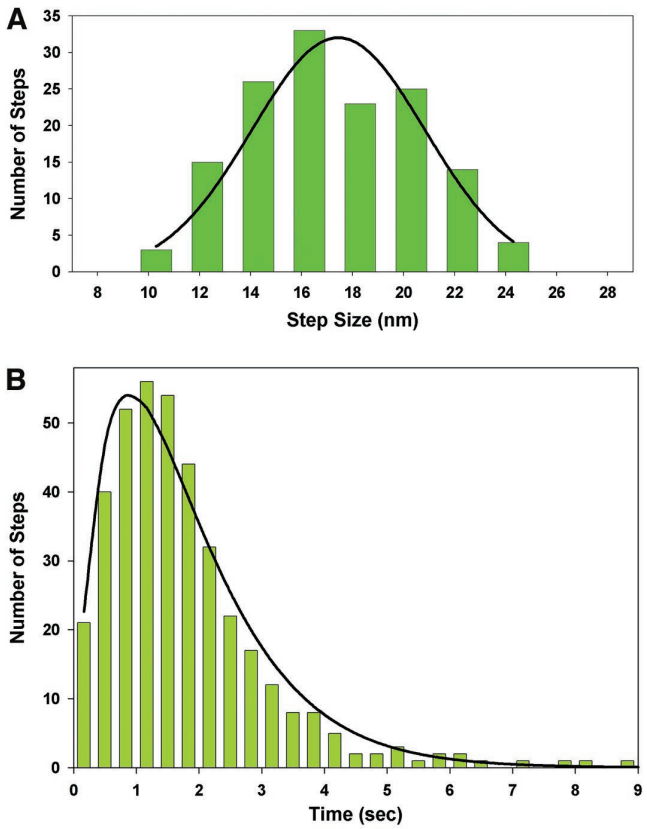

Figure 5: Experimentally determined histograms of A) fluorophore displacement distances and B) dwell times between displacements. From Yildiz, et al., *Science*, **303**, 676–678, 2004.