

# MCB137L/237L: Physical Biology of the Cell

## Spring 2024

### Homework 13 — Extra Credit

#### (Due 5/7/24 at 2:00pm)

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This homework draws from biological phenomena and physical models we explored throughout the semester. Note that this homework is for extra credit and that it is completely optional. As we are already going to drop your two lowest-scoring homework, if you complete this homework, we will use it to replace your third lowest-scoring homework. However, we will only do so if it actually *improves* your grade.

## 1 A minimal genetic switch

In class, we introduced how genetic switches can be constructed using two repressors that repress each other's gene expression. Here, we consider a simpler regulatory architecture that can also result in a genetic switch. Specifically, we will model the self-activation of an activator molecule. For this problem, you might find it useful, to review the phase portrait concept we described in class for the case of mRNA production and degradation, as well as the phase diagram of the logistic equation you had to draw earlier on in the semester.

Figure 1(A) presents a regulatory architecture, where an activator activates its own production. Figure 1(B) shows the states and weights for our model. In this problem, we will ignore mRNA and associate the rate indicated in the figure with the rate of protein production. Here, a promoter has two activator binding sites in its vicinity. In the absence of activators, or in the presence of only one activator, the rate of protein production is  $r_0$ . When both activators are bound, the rate is  $r$ . The activators bind with a dissociation constant  $K_d$  and interact with a cooperativity factor  $\omega = e^{-\beta\varepsilon_{int}}$ , where  $\varepsilon_{int}$  is the interaction energy between activators.  $A$  is the concentration of activator.

**(a)** Write down an equation describing the temporal evolution of the number of activators. Consider the rate of production stemming from the model shown in Figure 1, as well as a rate of protein degradation  $\gamma$ . Hint: Remember that the overall rate of production  $\langle r \rangle$  of a

system is given by

$$\langle r \rangle = \sum_i p_i r_i, \quad (1)$$

where  $p_i$  is the probability of the system being in state  $i$ , and  $r_i$  is the rate of production when the system is in that state.

- (b) Plot the phase diagram for this equation in order to find how many equilibria the system can support. Namely, plot the rates of production and degradation as a function of the activator concentration. Use  $K_d = 5 \text{ nM}$ ,  $\gamma = 0.1/\text{min}$ ,  $r_0 = 0.01 \text{ nM}/\text{min}$ , and  $r = 0.5 \text{ nM}/\text{min}$ . Make plots for  $\omega = 1$  and  $\omega = 10$ .
- (c) Draw vectors indicating the direction of the concentration change under your plots as we did in class for the mRNA production and degradation case, and as you explored in the homework in the context of the logistic equation. How many equilibrium points do you find? Indicate whether these points correspond to stable or unstable equilibria. You can review the concept of phase portraits by reading “Computational Exploration: Growth Curves and the Logistic Equation” on page 103 of PBoC2, paying special attention to vectors drawn on the lower part of Figure 3.10.
- (d) Solve the equation you derived in (a) for different initial conditions, and plot all of them on the same graph. Choose initial conditions that help illustrate how the system can converge to different levels of activator in steady state.

## 2 Uncovering phase separation in P-granule formation

In the paper by Brangwynne *et al.* (provided on the course website), the authors consider two mechanisms for the accumulation of P granules in the posterior end of the *C. elegans* embryo. First, they posit that P granules could migrate from the anterior end to the posterior end of the embryo. Second, they propose that anterior P granules could be preferentially disassembled or degraded.

- (a) Read their paper and write a one-paragraph summary of it. Make sure to explain the various hypotheses they considered and how they tested them.
- (b) In their Figure 4, they propose that, upon dissolution in the anterior end, the proteins that make up the P granules diffuse toward the posterior end to take part in granule formation at that location. Assume that these proteins have a reasonable diffusion constant, and estimate the time it takes for these molecules to diffuse throughout the embryo. How do these time scales compare to the overall rates of P granule formation?

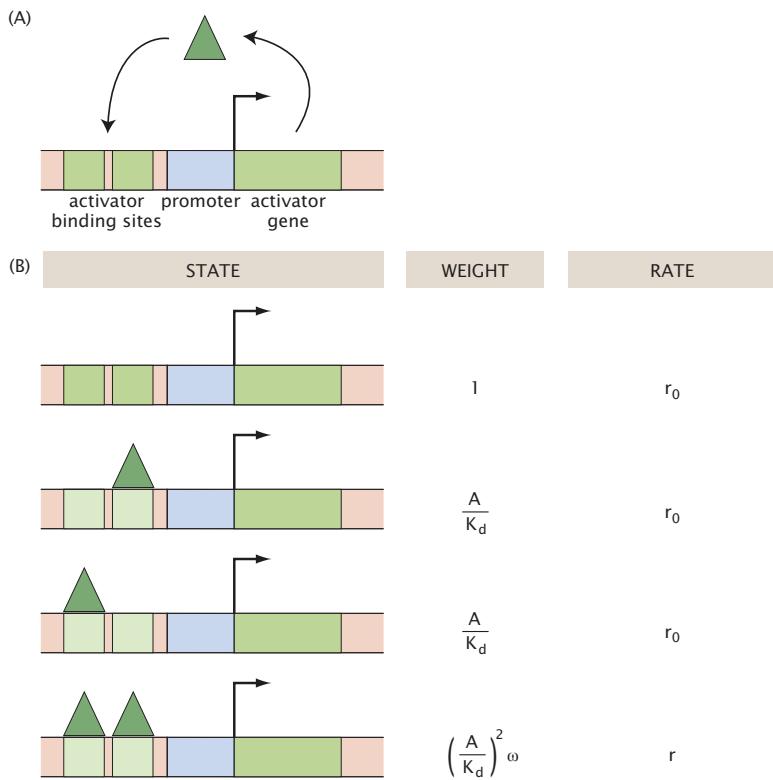


Figure 1: A simple autoactivation switch model. (A) Cartoon of the autoactivation switch. (B) States, weights and rates for the autoactivating genetic switch model.

### 3 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.

(b) In class, we posited that protein synthesis constitutes the largest source of ATP consumption, with 4 ATP equivalents required per amino acids incorporated. 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?

(c) 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.

## 4 Random walks and biological polymers

Physicists know how to solve just a handful of problems. Fortunately, many dissimilar phenomena in physics and biology alike can be mapped onto such problems for which we know a solution. Here, we explore the mathematical connection between diffusion and the spatial arrangement of polymers such as DNA, actin, and microtubules.

- (a) Read the introduction to Section 8.2 of PBoC (“Random Walk Models of Macromolecules View Them as Rigid Segments Connected by Hinges”) to learn more about how polymers can be thought of as chains of connected rigid segments. Pay close attention to Figures 8.1 and 8.2. Here, the Kuhn length  $a$  is defined as the length of the segments. Look up the Kuhn length for DNA, actin, and microtubules in order to get a feeling for these polymers. Note that you might find reference to the persistence length  $\xi_p = a/2$  instead of the Kuhn length.
- (b) Now, think of a polymer chain of  $N$  segments in 1D. As shown in Figure 8.3 of PBoC each segment can either be pointing to the right or to the left. Given  $n_R$  and  $n_L$  segments pointing to the right and left, respectively, the position of the end of the chain is given by  $L = (n_R - n_L)a$ . Map this problem onto the diffusion problem we solved in class where we calculated the  $\langle x \rangle$  and  $\langle x^2 \rangle$  of a random walker that start at the origin shown in Figure 2. To make this possible, note that each segment can be randomly pointing to the left or right. In particular, calculate  $\langle n_R - n_L \rangle$  and  $\langle (n_R - n_L)^2 \rangle$  and show that the size of the polymer is given by

$$\text{size} \approx \sqrt{\langle L^2 \rangle} = a\sqrt{N} \quad (2)$$

by repeating graphical the derivation we did in class.

- (c) Think of the size of the polymer you derived in (b) as the linear dimension of the blob the polymer will make on a surface such as shown in the figures below. Use the derived formula to estimate the genome length (in  $\mu\text{m}$  and bp) of the bacteriophage T2 shown in Figure 1.16 of PBoC and of the *E. coli* in Figure 8.5 of PBoC. How well did your estimate do?

All relevant figures from PBoC can also be found in Figures 3 and 4 below.

## 5 The French flag model

One of the most important and interesting ideas to come out of the idea of positional information contained in morphogen gradients was the so-called French flag model which we will explore here. This model posits that the Bicoid concentration dictates the position of the

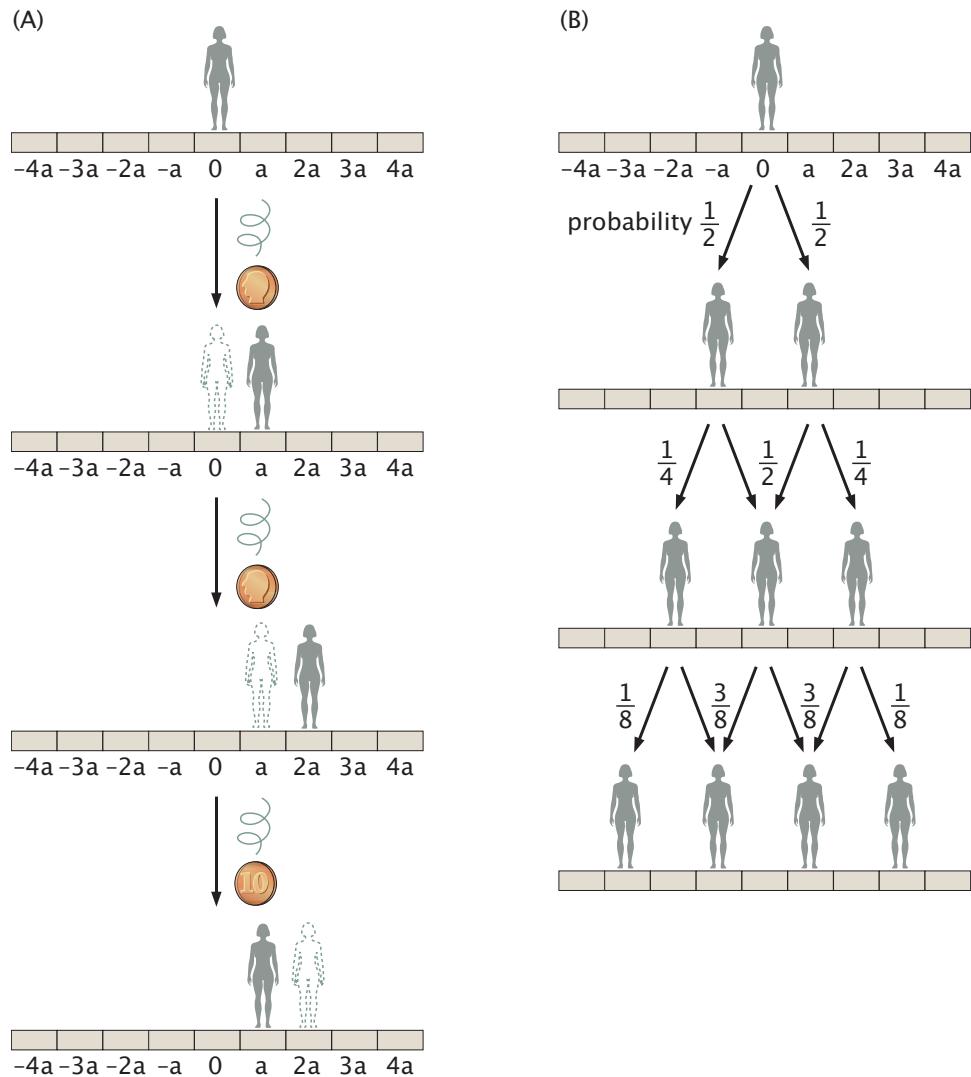
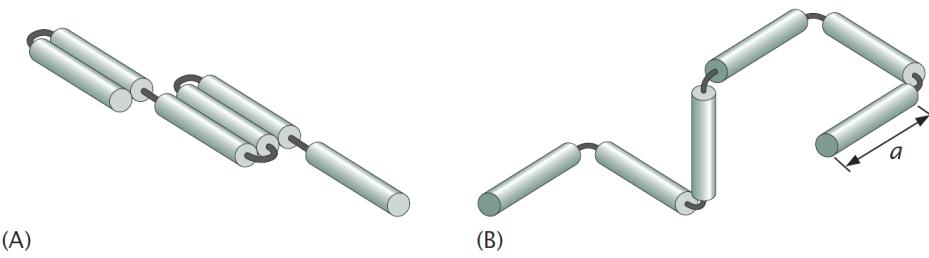
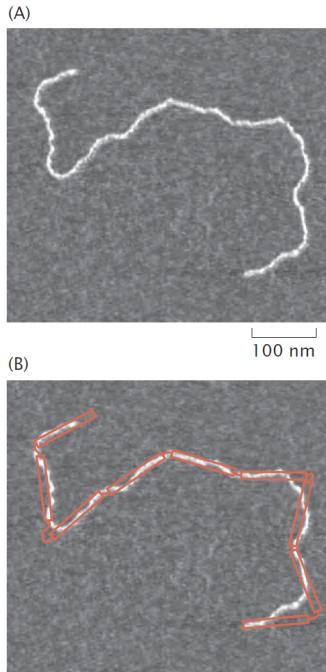


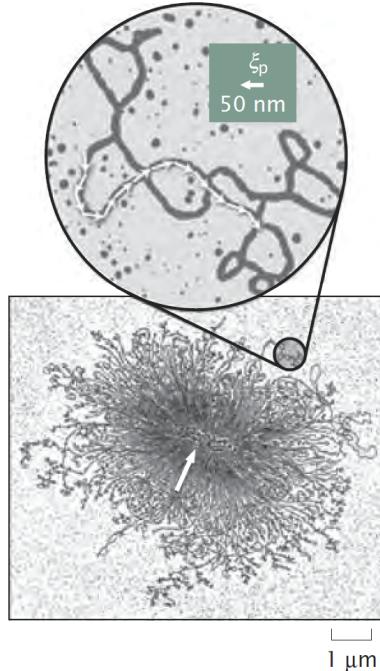
Figure 2: Coin flips and diffusion. (A) Stochastic “simulation” of a coin flipping process with the random walker stepping to the right when a heads is flipped and stepping to the left when a tails is flipped. (B) Scheme for calculating the probability of each and every possible outcome after a total of  $N$  steps.



**Figure 8.1:** Random walk model of a polymer. Schematic representation of (A) a one-dimensional random walk and (B) a three-dimensional random walk as an arrangement of linked segments of length  $a$ .



**Figure 8.2:** DNA as a random walk. (A) Structure of DNA on a surface as seen experimentally using atomic-force microscopy. (B) Representation of the DNA on a surface as a random walk. (Adapted from P. A. Wiggins et al., *Nat. Nanotech.* 1:37, 2006.)



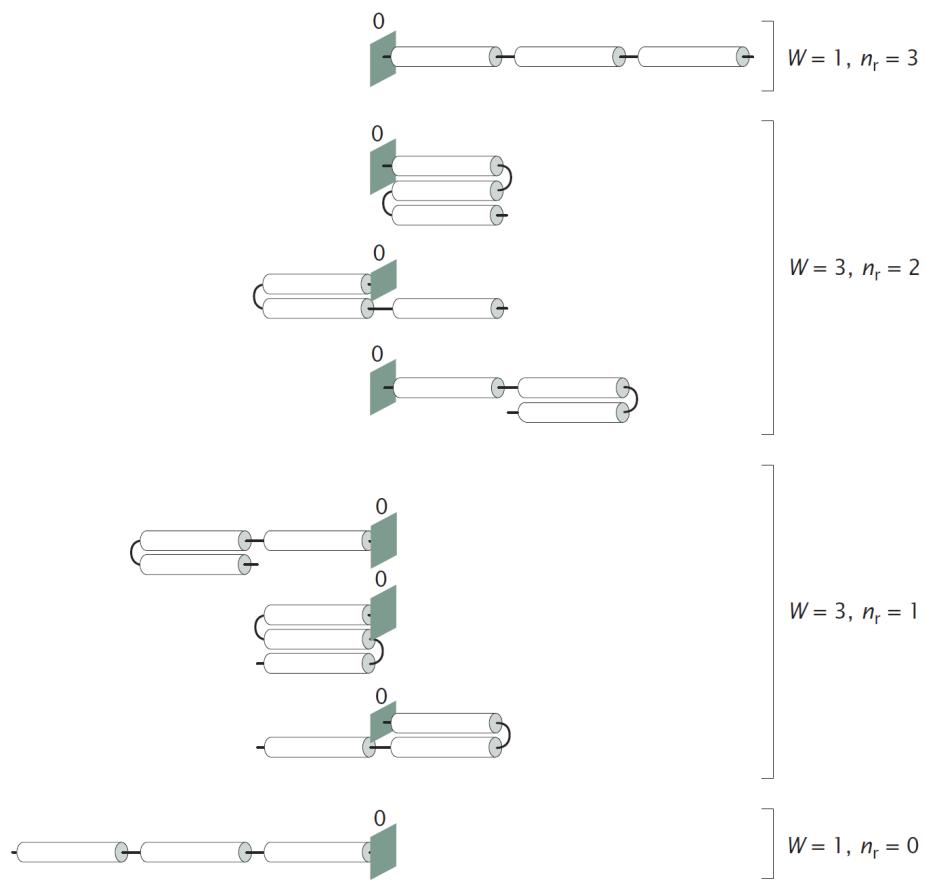
**Figure 8.5:** Illustration of the spatial extent of a bacterial genome that has escaped the bacterial cell. The expanded region in the figure shows a small segment of the DNA and has a series of arrows on the DNA, each of which has a length equal to the persistence length in order to give a sense of the scale over which the DNA is stiff. (Adapted from an original by Ruth Kavenoff.)



**Figure 1.16:** Electron microscopy image of a bacteriophage genome that has escaped its capsid. Simple arguments from polymer physics can be used to estimate the genomic size of the DNA by examining the physical size of the randomly spread DNA. We will perform these kinds of calculations in Chapter 8. (Adapted from G. Stent, *Molecular Biology of Bacterial Viruses*. W. H. Freeman, 1963.)

Figure 3: Figures 8.1, 8.2, 8.5 and 1.16 from PBoC.

cephalic furrow. As seen in Figure 5, the idea of the model is that boundaries in the embryo are determined by threshold values of the morphogen. The model predicts that, if the gene dosage of the morphogen gets changed, as seen in the mutant profile, the boundary will still occur at the same value of the morphogen. That hypothesis is enough to determine the shift in boundary position with gene dosage.



**Figure 8.3:** Random walk configurations. The schematic shows all of the allowed conformations of a polymer made up of three segments ( $2^3 = 8$  conformations) and their corresponding degeneracies.

Figure 4: Figure 8.3 from PBoC.

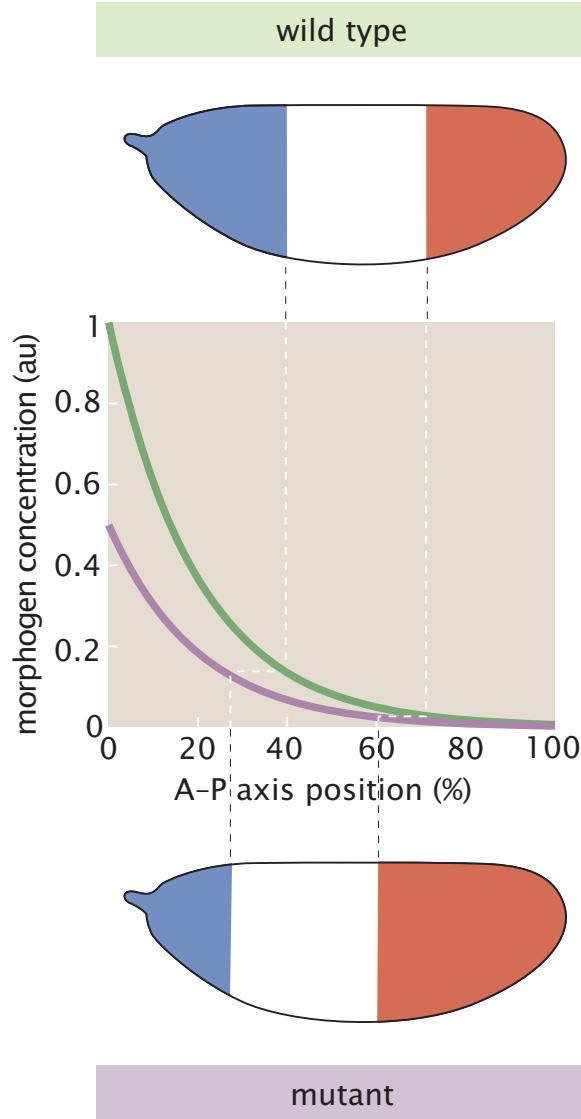


Figure 5: Concept of the French flag model.

To test this model, we will analyze several experiments (Nusslein-Vohlhard and Driever. and Liu *et al.*) where they measured cephalic furrow position as a function of different dosages of the *bicoid* gene in embryos. An exponential gradient of Bicoid is described by

$$Bcd(x, \lambda, \alpha, Bcd_0) = Bcd_0 \alpha e^{-x/\lambda}, \quad (3)$$

where  $x$  is the position along the embryo,  $Bcd_0$  is the Bicoid concentration at  $x = 0$ ,  $\lambda$  is the decay constant of the gradient and  $\alpha$  is the Bicoid dosage, with  $\alpha = 1$  corresponding to the wild-type.

- (a) Work out a model that predicts the position of the cephalic furrow  $x_{new}$  as a function of the gene dosage  $\alpha$ , the morphogen gradient decay length  $\lambda$  and the position of the wild-type cephalic furrow,  $x_{CF}$ .

**(b)** Note that, given a measured  $x_{CF} \approx 32\%$  of the embryo length, your model has no free parameters. Compare the prediction from your model with the data for  $x_{new}$  vs.  $\alpha$  obtained by Nusslein-Vohlhard, and by Driever and Liu *et al.* (provided on the course website). Comment on how well your prediction matches the data that is provided with the homework. What could be going on?