MCB137L/237L: Physical Biology of the Cell
Spring 2024
Homework 9:
(Due 4/2/24 at 2:00pm)

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"Biology is catching up" - PAM Dirac

1 Phase Separation from a Dynamical Systems Perspective Revisited

In class we introduced an approach for thinking about phase separation from the perspective of dynamical systems. Specifically, as shown in Figure 1, we assumed that individual FIB-1 molecules joined the nucleolus at a rate k_{on} and left at a rate k_{off}. In that model, we did not assume any dependence of these rates with the concentration.

In this problem, we explore an alternative model that does not assume k_{on} or k_{off} to be constant. Specifically, as shown in Figure 2, we will assume that the on rate is proportional to the surface area of the condensate. To put this in other words, the larger the condensate, the more likely it is for a molecule to enter it. Further, we also assume that the off rate is proportional to the surface area of the condensates: the more surface area the more likely

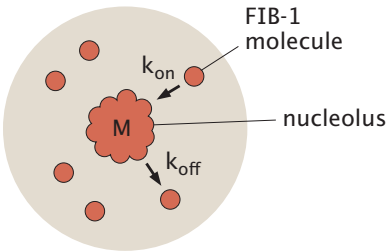

Figure 1: Simple model of nucleolar assembly. A nucleus of volume V contains a total of N FIB-1 molecules, M of which are assembled into the nucleolus. Single FIB-1 molecules are incorporated into the nucleolus at a rate k_{on}, and are separated from the nucleolus at a rate k_{off}.

that a molecule will leave the condensate.

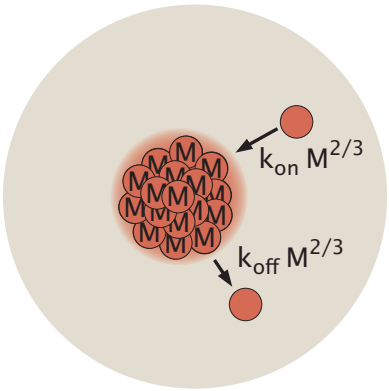

Figure 2: Model of phase separation accounting for the size of the condensate. In this model, the rates of incoporation and loss of molecules in the condensate is proportional to the surface arear of the condensate.

(a) Given our model, explain why the on and off rates are given by $k_{on}M^{2/3}$ and $k_{off}M^{2/3}$, respectively. Here k_{on} and k_{off} are constants.

(b) Show that the dynamical equation describing the number fo molecules in the condensate is given by

$$\frac{dM}{dt} = (N - M)k_{on}M^{2/3} - k_{off}M^{2/3}, \tag{1}$$

where N is the total number of molecules in the nucleus. Make sure to explain where the $(N - M)$ term is coming from.

(c) Determine the conditions necessary for phase separation to take place.

(d) Plot the rate of molecule incorporation and loss as a function of M for the case where phase separation can take place as well as for the scenario where phase separation cannot take place. Make sure to incorporate vectors under the plot that indicate the dynamics of the system as we have done in class.

(e) Plot the number of molecules within the condensate M as a function of N for the case where phase separation is possible. Also, plot the number of free molecules $N - M$ as a function of N in this same regime. Make sure to point out what happens to the amount of free molecules once phase separation ensues.

2 Ligand-Receptor Problem from the Perspective of Statistical Mechanics

In Homework 4, we calculated ligand-receptor binding from the perspective of rate equations and dissociation constants. In this problem we explore a third route to compute the probability of a receptor being bound by a ligand based on statistical mechanics. Note that you can complement this problems by reading the paper "A First Exposure to Statistical Mechanics for Life Scientists: Applications to Binding" (Garcia2007b) on the course website.

(a) Imitate the statistical mechanics protocol given in class by showing the states, energies, multiplicities and weights for a lattice consisting of Ω lattice sites and L ligands. Find an expression for p_{bound} in terms of the difference in energy of a ligand when in solution, ε_{sol} and the energy when the ligand is bound to the receptor, ε_b.

(b) Consider that the lattice sites in our lattice model (Figure 1 of Homework 4) have size v and hence that the concentration is $[L] = L/\Omega v$ and use that insight to arrive at an expression for the dissociation constant in terms of the microscopic parameters ε_{sol} and ε_b. Do this by comparing the results of this part of the problem with your result from Homework 4

3 Mutation Per Generation in Humans

Comparing genetic sequences has served as a useful tool for determining how various organisms are related to each other. With the advent of the "genomic era," we no longer have to infer how living organisms are related to each other based on morphological traits alone. In this problem, we will begin to get a sense of the time scales over which mutations accumulate in genetic sequences and how we can use these mutations as a molecular clocks for determining the relationships between various organisms.

In this problem, we are ultimately interested in estimating the total number of mutations that are passed on in each human generation. As a first step, we must estimate the number of mutations that accumulate in a single cell division.

(a) Given that the human genome is 3 billion basepairs long and is replicated with an incredible fidelity of only one error in every 10^{10} basepairs per replication, how many mutations do you expect to see after one genome duplication?

With this number of mutations per genome duplication in hand, we can next tackle how many mutations are passed on by a mother and a father. Recall that while many mutations may occur in a given human, only those that accumulate in the gametes (egg and sperm) will actually be passed on. To determine the number of mutations that we expect to be passed on, we will need to consider the formation of the egg and the sperm separately as males and

females have different developmental pathways regarding gametogenesis (see Figure 2).

As a primer for thinking about gametogenesis, let's briefly review the difference between mitosis and meiosis. Mitosis is the process by which a somatic cell duplicates its genome and then divides into two cells. Thus in a human, mitosis yields two cells with 46 chromosomes each. Meiosis, however, is the process by which a cell duplicates its genome and then proceeds to undergo two cell divisions, ultimately resulting in four cells with 23 chromosomes. This means that each round of mitosis requires one genome duplication and each round of meiosis requires one genome duplication (despite having two cell divisions).

In humans, females are born with all of their eggs nearly fully developed and they produce no new egg cells throughout the rest of their life. As illustrated in the top half of Figure 2, every developed egg is the result of 22 rounds of mitosis and 1 round of meiosis, yielding a total of 23 genome replications. This means that every egg a woman produces has undergone 23 genome replications regardless of a woman's age.

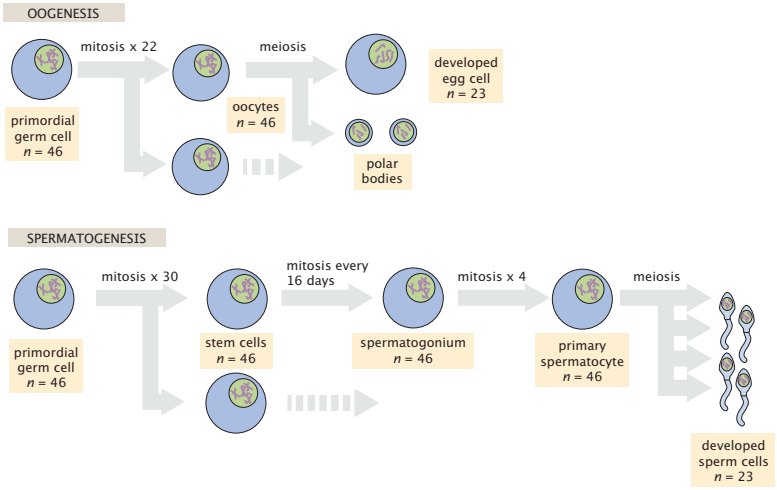

Figure 3: Schematic of oogenesis and spermatogenesis in humans. n refers to the number of chromosomes, where somatic cells have 46 and gametes have 23. For simplicity, the dashed arrows indicate the lineages of cells that we do not follow.

(b) Given the 23 genome duplications that occur in the process of forming an egg, how many mutations do you expect a woman to pass on to her children?

By contrast, spermatogenesis occurs continually throughout a male's lifetime upon reaching sexual maturity (i.e. puberty). At a bare minimum, a developed sperm cell has undergone 34 rounds of mitosis (30 leading to the formation of the stem cell and 4 after the stem cell) and 1 round of meiosis. But there are also additional rounds of mitosis to take into account as the result of the stem cells continually dividing to maintain the sperm supply. With these stem cells dividing every 16 days after puberty, the number of genome duplications to make a man's sperm is dependent on the age of the man.

(c) How many genome replications have occurred to make a "typical" man's sperm? In this context, we consider that a "typical" male hits puberty at 15 and reproduces at 30 years old.

(d) Given your answer in (c), how many mutations do you expect this "typical" man to pass on to his children?

We have now estimated the total number of mutations that we expect the mother and the father to contribute, allowing us to determine the total number of mutations per human offspring.

(e) What is the total number of mutations we expect to accumulate in a human offspring? What are the relative effects of the mother and the father in this estimate?

(f) Make a plot of the number of mutations accumulated in the gametes as function of age for males and females. Make sure to graph the number of mutations in the egg and the sperm on the same plot to better compare their relative effects.

4 Ion Channels and Statistical Mechanics

In this problem, we will derive a mathematical description of the current passing through a voltage-gated ion channel. To model this channel, we assume that it can exist in an open or closed configuration as shown in Figure 4A. The thermal fluctuations in the cell result in the channel switching between these states over time as presented in Figure 4B. Figure 4C shows how these fluctuations in channel state can be directly read out from the current flowing through the channel.

(a) Use the statistical mechanics protocol (i.e. calculating the states and weights of the system) to calculate the probability of the channel being in the open state, p_{open}. Assume that the open state has an energy ε_{open}, and that the energy of the closed state is ε_{closed}.

(b) Plot p_{open} as a function of $\Delta\varepsilon = \varepsilon_{open} - \varepsilon_{closed}$. Explain what happens in the limits $\varepsilon_{open} \ll \varepsilon_{closed}$ and $\varepsilon_{open} \gg \varepsilon_{closed}$. What significance does $\Delta\varepsilon = 0$ have for p_{open}?

In a simple model of a voltage-gated ion channel, $\Delta\varepsilon = q(V^* - V)$. Here, V is the voltage applied to the membrane and q is the effective gating charge, which describes the movement of charges along the membrane as the channel configuration changes. You can learn more about this model in section 17.3.1 of PBoC2.

(c) What is the significance of V^*? Namely, what happens to the probability of being open when $V = V^*$.

(d) On the website, you will find measurements of p_{open} vs. V for a sodium-gated ion channel. Write your expression for p_{open} as a function of V instead of as a function of $\Delta\varepsilon$. Estimate

V^* from the data using what you learned in (c). Now that you have V^*, to estimate q, make a plot where you overlay the data and the model prediction for three different values of q corresponding to 1, 3 or 5 electron charges (note that q is positive, so here we are talking about the *absolute value* of the electron charge).

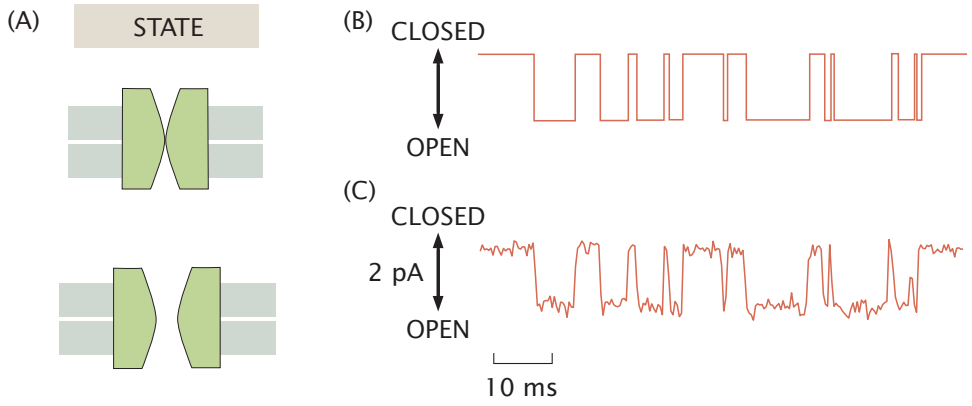

Figure 4: Current through an ion channels. (A) The ion channel can exist in a closed or open configuration, (B) fluctuating in time between these two states. (C) The current flowing through the channel is directly related to the state of the channel.