MCB137L/237L: Physical Biology of the Cell
Spring 2020
Homework 7: Phase Separation and Statistical Mechanics
(Due 4/2/20 at 3:30pm)

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"The quantum physicist Richard Feynman once gave a lecture on color vision in Caltech's Beckman Auditorium. He explained the molecular events that take place in the human eye and brain to show us red, yellow, green, indigo, and blue. This chain of reactions was one of the early discoveries of molecular biology, and fascinated Feynman. 'Yeah,' someone in the audience said, 'but what is really happening in the mind when you see the color red?' And Feynman replied, 'We scientists have a way of dealing with such problems. We ignore them, temporarily.' " - Jonathan Weiner in *Time, Love, Memory.*

1 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?

2 Free energy minimization and phase separation

In class, we derived the free energy for a simple model where red and blue molecules are mixed. In this problem we repeat some of those derivations and explore how the choice of

parameters can dictate whether the solution will phase separate or not. In performing these calculations, make sure to explain each one of your steps.

(a) Calculate the free energy of the system G as a function of $\phi = \frac{N_R}{N_R+N_B}$, where N_R and N_B are the number of red and blue particles, respectively. Specifically, repeat our calculations for the entropy and interaction energy between particles.

(b) Using Python, plot the free energy as a function of ϕ for different temperatures. Assume $V_{RR} = V_{BB} = -2\ k_B T_0$ and $V_{RB} = 0.5\ k_B T_0$, where $T_0 = 37.0°C$. Explain what features in the free energy plot indicate that the solution will phase separate, and what determines the concentrations of red molecules ϕ' and ϕ'' in each phase.

(c) Posit that the solution will phase separate when the free energy becomes concave such that $\frac{d^2G}{d\phi^2} < 0$ and show that this condition demands that

$$\frac{Tk_B}{\phi(1-\phi)} < -\chi, \tag{1}$$

where $\chi = V_{RR} - 2V_{RB} + V_{BB}$. Explain how χ captures the competition between attraction of like molecules and repulsion between red and blue molecules.

(d) By enforcing the inequality you derived above, show how changing temperature, ϕ, and χ can be used to determine whether the solution will phase separate. To make this possible, use Python to make plots such as the one shown in class for the two terms in Equation 1 as a function of ϕ for different values of T and of χ.

3 Ligand-receptor problem from the perspective of statistical mechanics

In Homework 6, 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.

(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 6) 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 6.

4 Mutation correlation and physical proximity on the gene

(a) Read the section "Flies and the Rise of Modern Genetics" starting on page 170 of PBoC2.

(b) Do problem 4.4 from PBoC2.

4.4 Mutation correlation and physical proximity on the gene

In Section 4.6.1, we briefly described Sturtevant's analysis of mutant flies that culminated in the generation of the first chromosome map. In Table 4.2, we show the crossover data associated with the different mutations that he used to draw the map. A crossover refers to a chromosomal rearrangement in which parts of two chromosomes exchange DNA. An illustration of the process is shown in Figure 4.26. The six factors looked at by Sturtevant are B, C, O, P, R, and M. Flies recessive in B, the black factor, have a yellow body color. Factors C and O are completely linked, they always go together and flies recessive in both of these factors have white eyes. A fly recessive in factor P has vermilion eyes instead of the ordinary red eyes. Finally, flies recessive in R have rudimentary wings and those recessive in M have miniature wings. For example, the fraction of flies that presented a crossover of the B and P factors is denoted as BP. Assume that the frequency of recombination is proportional to the distance between loci on the chromosome.

Reproduce Sturtevant's conclusions by drawing your own map using the first seven data points from Table 4.2.

Keep in mind that shorter "distances" are more reliable than longer ones because the latter are more prone to double crossings. Are distances additive? For example, can you predict the distance between B and P from looking at the distances B(C,O) and (C,O)P? What is the interpretation of the two last data points from Table 4.2?

Table 4.2: Fraction of crossovers of six sex-linked factors in *Drosophila.* (Adapted from A. H. Sturtevant, *J. Exp. Zool.* 14:43, 1913.)

Factors	Fraction of crossovers
BR	115/324
B(C,O)	214/21736
(C,O)P	471/1584
(C,O)R	2062/6116
(C,O)M	406/898
PR	17/573
PM	109/458
BP	1464/4551
BM	260/693

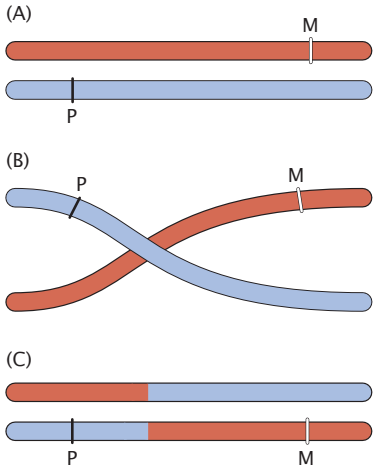

Figure 4.26: Crossing over of chromosomes. (A) Chromosomes before crossing over showing two loci labeled P and M. (B) Illustration of the crossing-over event. (C) Chromosomes after crossover.

Figure 1: Problem 4.4 from PBoC.