APh161: The Physics of Biological Structure and Function
Homework 4
Due Date: Thursday, February 10, 2005

"An ounce of application is worth a ton of abstraction." - Booker T. Washington

Reading: Chap. 4 of Schleif and chap. 2 of Boal.

1. Poster Abstracts.

Please submit by email to both me and the TAs (Rizal and Hernan) a one paragraph statement of what your poster will be about. Recall that the poster has to be built up in the spirit of the calculations I have done in class. That is, you need to introduce some biological phenomenon and then use quantitative arguments to describe it.

NOTE: please put problems 2 and 3 on separate sheets of paper. This will make grading easier.

2. Nucleosome Formation and Assembly

The goal of this problem is for you to reexamine the ideas developed in class concerning the assembly and accessibility of the nucleosome.

(a) Repeat the derivation given in class and arrive at an expression for the free energy of formation of nucleosomes. Make sure that you explain the qualitative features of the calculation and that you identify the numerical outcome of the analysis (please report your energies in units of k_BT). In particular, comment on the way in which you handle the elastic and interaction effects and rationalize the energy per length that you assign to each of these effects. Also, instead of pretending that the DNA wraps around the nucleosome fully two times, use the more precise description involving 146 base pairs of wrapped DNA leading to 1.75 wraps around the histones. A second refinement is to include the helical pitch of the DNA as it wraps around the

nucleosome - here, just make an estimate to show that this refinement is not very important. (Hint: to do this you will need to remember that the bending energy is of the form

$$E_{bend} = \frac{\xi_p k_B T}{2} \int ds \kappa(s)^2, \tag{1}$$

where $\kappa(s)$ is the curvature.) In addition, take a look at papers describing the structure of the nucleosome (such as Nature, **389**, 251 (1997); Nature **423**, 145 (2003); J. Mol. Bio. **319**, 1097 (2002)) and make sure that you characterize the structural features of the nucleosome that you use in your model.

(b) In class, we examined the experiments of Polach and Widom (J. Mol. Biol., **254**, 130, (1996)) which examined the equilibrium accessibility of different sites within the nucleosome as a function of the distance within the nucleosome that the DNA binding site is buried. In this part of the problem, you will reexamine the derivation of the equilibrium accessibility and explicitly compare your results with those of Polach and Widom. As I did in class, derive expressions for the fractional coverage of different sites as a function of how deeply they are buried in the nucleosome and explain in detail the arguments leading to the result. Next, derive the equilibrium constant we call $K_{eq}^{conf}(x)$ and compare your results explicitly to those from Polach and Widom. Make sure you are careful in describing the logic of reconciling the microscopic model and the description in terms of equilibrium constants. As part of the procedure to compare to Polach and Widom, you will have to fit the adhesive energy γ that I used to characterize the histone-DNA interactions.

3. DNA Packing in Bacteriophage.

(a) In class we discussed bacteriophage such as lambda phage, T4, T7, P22 and phi29. In this part of the problem, you will make an estimate of the spacing between the DNA strands when packed in their ordered arrangement as a function of the length packed in the capsid using strictly geometric arguments. In the second part of the problem, you will have a chance to refine this description by including the elastic and interaction contributions to the overall free energy budget for packing. For simplicity, consider a spherical capsid

(rather than the faceted icosahedral capsids associated with real viruses). In particular, let's model lambda phage as a spherical capsid with a radius of 27nm. It has roughly a 48,000 bp genome. How long is that DNA? When the capsid is nearly filled with DNA, the repulsive interactions between neighboring DNA segments make the DNA try to be as far apart as possible. Your objective is to compute the spacing between the segments on the assumption that the DNA has adopted a locally hexagonal arrangment (when viewed head on) and that the lattice parameter of this spacing is as large as it can be. In particular, find an analytic formula for the spacing of the DNA as a function of the length L of DNA packed and the radius of the capsid. By examining mutants of lambda phage, Earnshaw and Harrison (Nature, **268**, 598 (1977)) were able to measure the spacing as a function of the length of DNA packed and find that at 78% filling, the spacing is 26.2$\mathring{A}$, when 88% full, the spacing is 25.1$\mathring{A}$, when 100% full, the spacing is 24.0$\mathring{A}$ and when overfilled to 105%, the spacing is 23.5 $\mathring{A}$. (If you are feeling ambitious, look at Cerritelli *et al.* Cell, **91**, 271 (1997), who performed similar measurements on T7). Make a fit of these data points for lambda to the scaling result you obtained.

b) In class, I worked out the elasticity theory for the packing of DNA in a cylindrical capsid. Repeat that calculation in the more realistic case of a spherical capsid (with dimensions chosen to correspond to λ phage). Obtain both the elastic energy and the elastic contribution to the force vs length packed curve.

c) My derivation in class was based on the approximation that the DNA was packed as a series of hoops. In fact, since the DNA is one long molecule, it is actually packed as a helix. Compute the radius of curvature for a helix (i.e. do the mathematics problem and show your result) and then use this curvature (rather than the hoop approximation) and compute the elastic energy for a cylindrical capsid and compare to the result I got in class. Comment on the limits and validity of ignoring the correction due to helical pitch. The main point of this part of the problem is for you to decide whether or not you buy the idea of abandoning the helical pitch of the packed DNA and treating everything just as circular hoops.

d) Finally, repeat the derivation that I did in class for the total free energy

of packing for a cylindrical capsid, but now for a spherical approximation to the phi29 capsid used in the Bustamante experiment. Use Mathematica or Maple or whatever scheme you like to solve for the d-spacing and the force for the cases in which 3000, 4000, 5000, 6000 and 6600 nm worth of DNA are packed. The parameters you will need to use are: $\xi_p = 50nm$, $k_BT = 4.1$ pN nm, $F_0 = 225,500\text{pN/nm}^2$, $c = .27nm$ and R =22 nm. This choice of the radius of the spherical capsid guarantees that the internal volume of our model capsid will have the same dimensions as the real phi29 capsid. Make a plot in which you explicitly compare your force vs percent packed curve to the data of Bustamante.