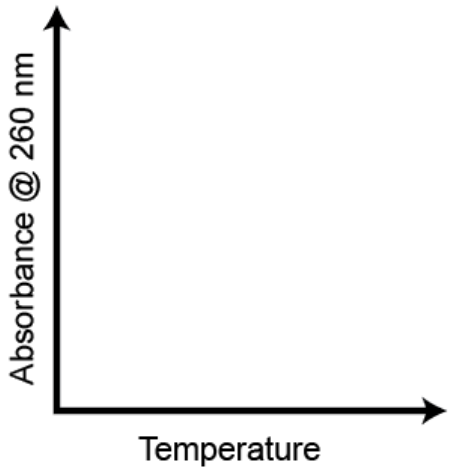
DNA Folding/Unfolding

1. DNA and RNA strongly absorb UV light. What part of these molecules is primarily responsible for this absorbance? Why did you choose this region?

2. The hyperchromic effect is used to study DNA melting. This is a general phenomenon that explains the difference in absorbance between folded and unfolded DNA and RNA molecules.
    a. Which absorbs light more strongly: folded or unfolded nucleic acids? Why did you pick this? Don't spend a lot of time on this in class, but be thinking about explanations behind physical observations that we make.

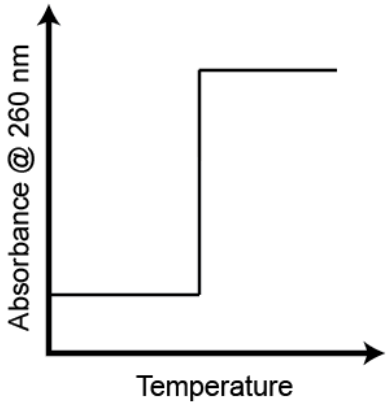
    b. Based on your answer above, predict what a DNA melting curve would look like. Melting curves monitor the unfolding of DNA as the temperature is increased.



    c. Mark your diagram with the melting temperature – this is the temp when 50% of the DNA is unfolded.
    d. Check out figure 24-22 in your book. Does the shape of the curve agree with your prediction?

    e. The sigmoidal shape ("S-shape") of this curve is very meaningful. What does this curve shape tell us about the melting of DNA?

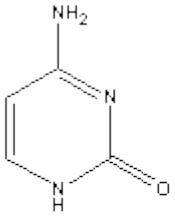
    f. What would a step-shaped melting curve mean?



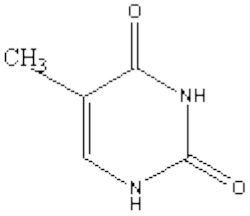
This exercise is going to help you understand the difference between the hydrophobic effect and pi stacking in DNA and consider the thermodynamic forces that stabilize DNA.

H-bonding

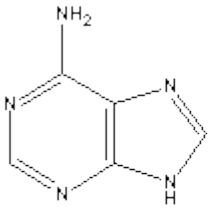
3. Below are the structures of each of the four bases in DNA.
    a. Identify all parts of the Watson and Crick face that will H-bond with water.
    b. Identify all parts of the Watson and Crick face that will H-bond with another base when DNA folds.
    c. Is there a difference between the number of H-bonds that will be present in folded and unfolded DNA?
    d. Based on your answers, do you think that H-bonding is an important part of the energy that helps DNA fold?



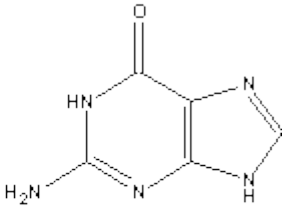
Cytosine



Thymine



Adenine

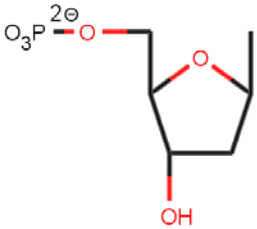


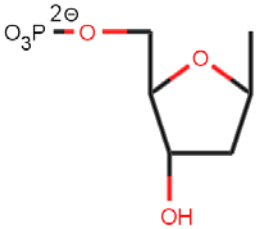
Guanine

Pi Stacking

4. Summarize the hydrophobic effect. What are the two steps that need to be considered to understand the enthalpy/entropy relationship? Why is water such an important part of the phenomenon?

5. Complete the structures of dAMP and dTMP.





a. On the structures above, identify all regions that will become shielded from solvent during DNA folding.

b. Are the regions that you identified above hydrophobic?

c. Can water interact with these regions using any of the common intermolecular forces? If so, which?

d. How would the solvent reorganization be different than what we discovered in the hydrophobic effect?

e. Do you expect the process of DNA folding to be driven primarily by entropy or enthalpy? Explain your choice.

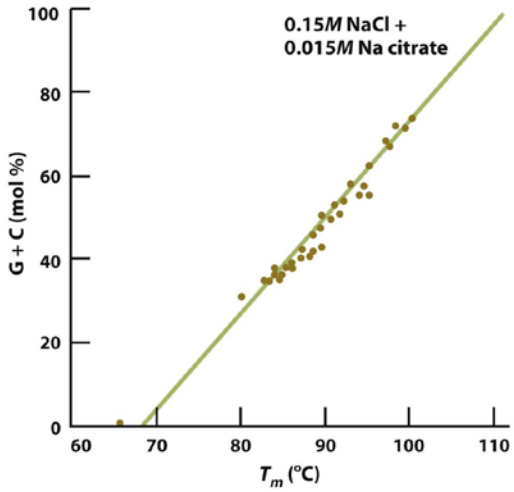
f. Based on your answers to the above questions, is DNA folding endothermic or exothermic?

g. Recalling that heat is a product in an exothermic reaction and a reactant for an endothermic process, can you use Le Chatelier's Principle to explain why DNA melts as the temperature increases.

A:T vs. G:C in DNA Stability

6. Summarize what this figure tells us about DNA stability.

7. As we figured out above, H-bonds are not particularly important in the energy that stabilizes DNA/RNA structures. Based on this, propose a reason that increasing the G-C content of DNA increases the melting temperature.



8. Check out Table 24-2 from your book. Does this data support your proposed reason? If not, use the information in this table to propose a reason that GC content stabilizes DNA structure.