Biology 5357

Chemistry & Physics of Biomolecules

Examination #2

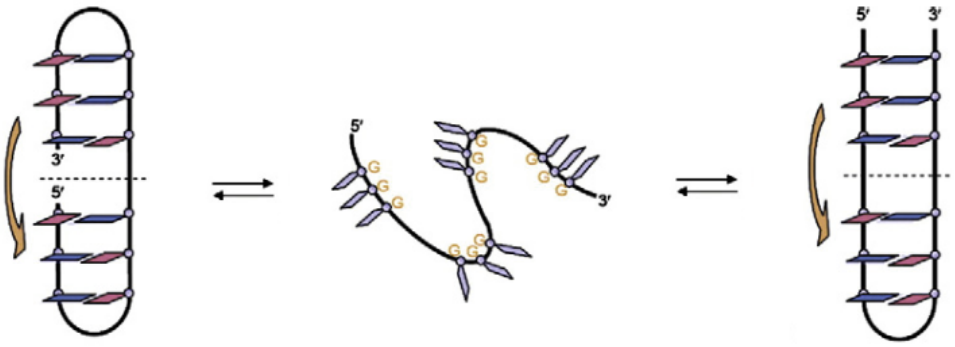
Nucleic Acids Module

November 5, 2021

Name: _____

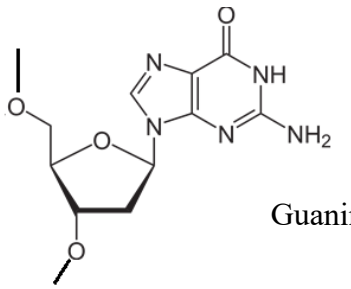
Question 1. (8 points) Explain why there is a preference for nucleosome core particles to form from oligomers of $(A/T)_3NN(G/C)_3NN$ (TG motifs), where X/Y indicates either nucleotide. Recall that a nucleosome core particle is an assembly of two each of H2A, H2B, H3 and H4 histone proteins around which about 150 bp of DNA wraps.

Question 2. (8 points; A & B=4 points each) In the initial step in the proposed pathway to G quadruplexes formed from the human telomeric sequence $(GGGTTA)_n$, two types of hairpin are proposed to form, a long hairpin and a double hairpin. In the figure only the G's are shown, and are linked by TTA.



(A) Write simple expressions for the free energy of each structure in terms of base pair stacks and loop free energies. What would the free energy difference between the two structures be principally due to?

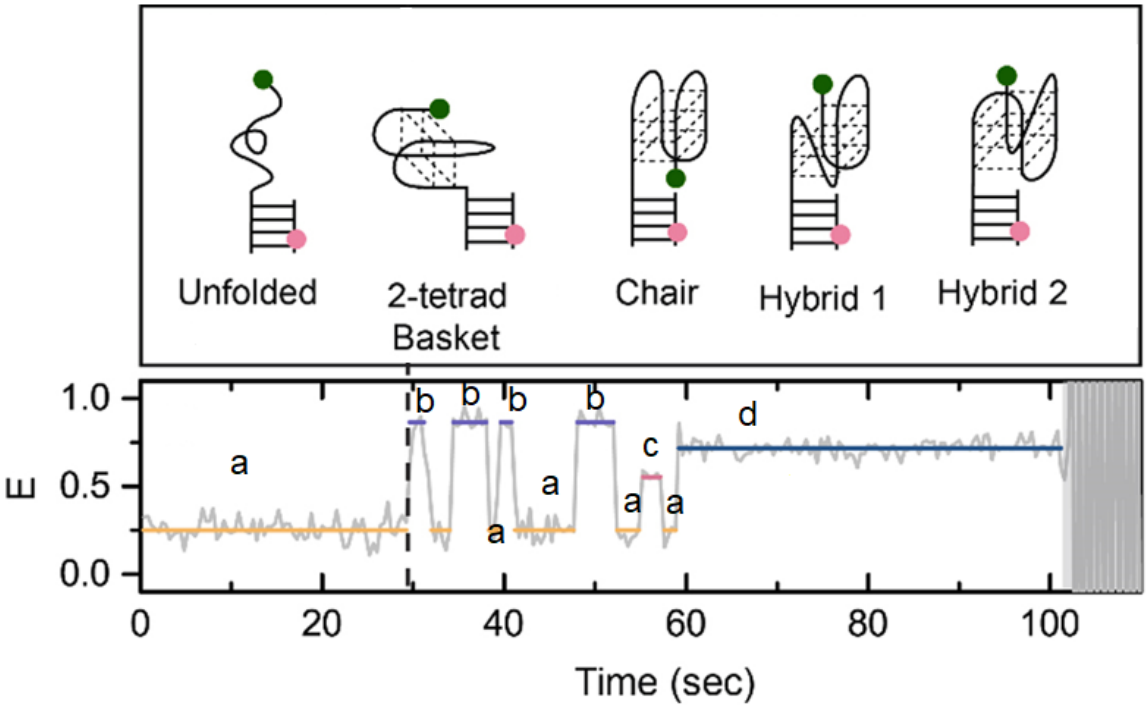
(B) If the hairpins were to form from G's in their most stable anti-glycosyl conformation, what type of G-G base pair would be involved. Sketch the base pair and H-bonding interactions which would result in an antiparallel duplex. (The structure of G is given below. You only need to show the base portion in your answer.)



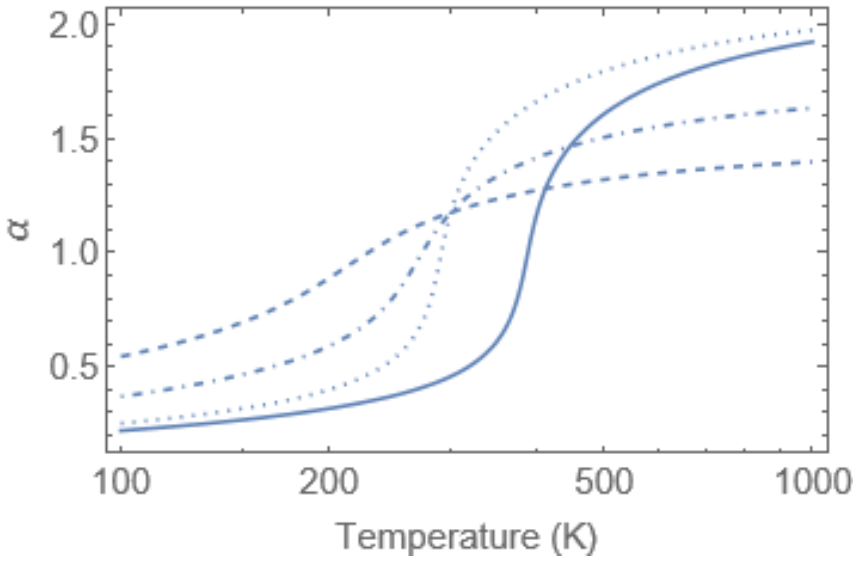
Guanine Nucleoside

Question 3. (8 points) In the proposed recognition codes for zinc fingers, A is most often recognized by either asparagine (Asn, N) or glutamine (Gln, Q). Draw the structure of an A-T base pair, and also draw how the A could be recognized by the side chain of either asparagine or glutamine.

Question 4. (8 points) The following figure shows schematic drawings of various conformations of a human telomere sequence with donor and acceptor fluorophores, and below, the fluorescence efficiency observed in a FRET experiment for a single molecule as a function of time before and after the addition of potassium ion at 29 seconds to initiate folding. Assign conformations to the species giving rise to the FRET efficiencies **a**, **b**, **c**, and **d**. What does the trace suggest about how one conformation converts to another, and which is the most stable species?



Question 5. (16 points; A-D=4 points each) Consider the plot shown below depicting the change of alpha (α) *vs*. temperature.



(A) Can you tell which of the curves (continuous, dashed, dotted, dot-dashed) correspond to the polymers of length 10, 100 and 1000?

(B) Can you quantify the theta (θ) temperature of each polymer?

(C) Can you identify the regimes of good and poor solvent, and indicate the corresponding exponent?

(D) As a first approximation, what is the ratio of the mean square end-to-end distance for the polymers of length 100 and 1000, assuming they have the same Kuhn segment length in the three different solvent regimes?

Question 6. (12 points; A-C=4 points each) The sequence of a disordered protein is 120 amino acids long. Remember the Cα-Cα distance is 0.38 nm (3.8 Angstroms) and the persistence length is 0.5 nm.

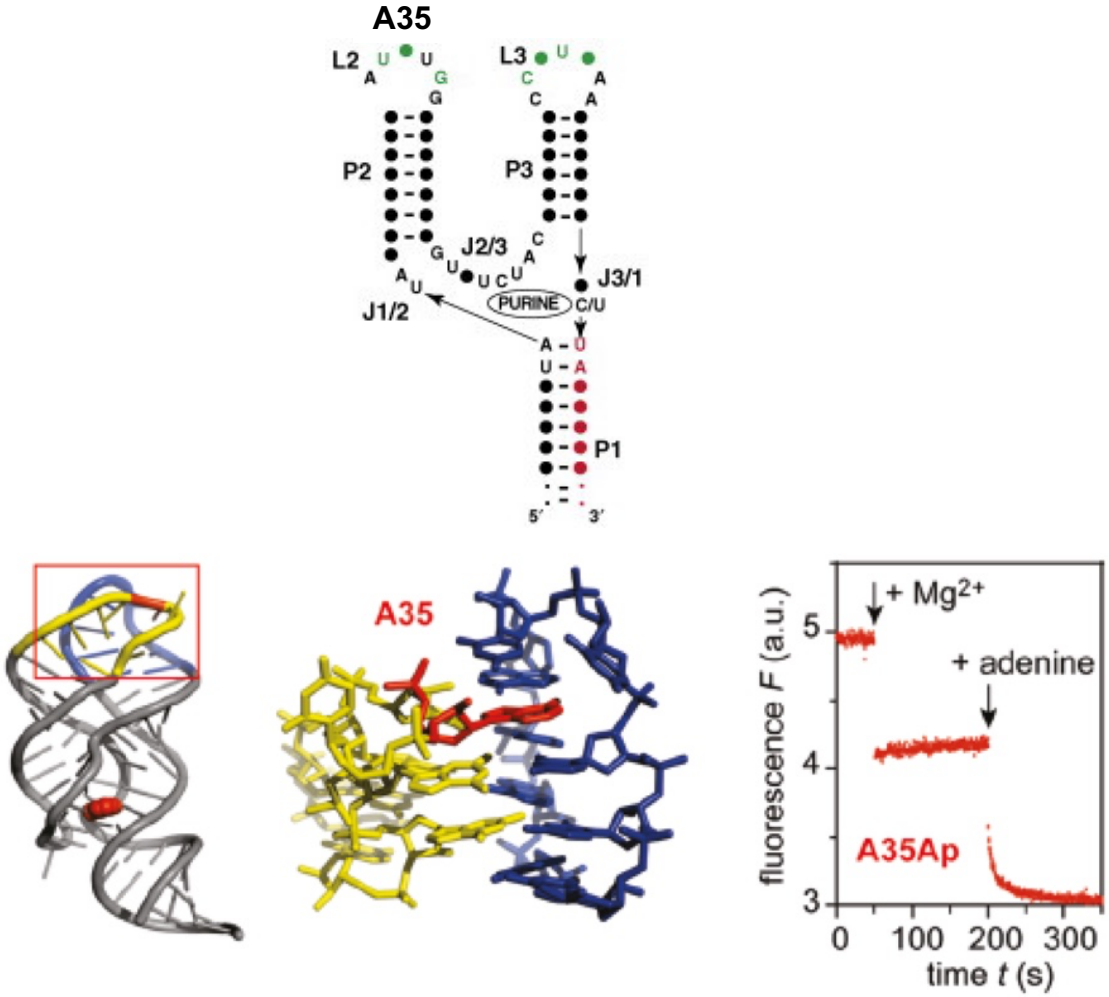
(A) Define and compute the contour length.

(B) Define and compute the number and length of Kuhn segments.

(C) Write the equation for the distribution of the end-to-end distances and radius of gyration.

Question 7. (10 points; A=6 pts, B=4pts)

The nucleobase 2-aminopurine (2AP) is fluorescent, but its fluorescence is quenched by base stacking. In this purine riboswitch, A35 is replaced by 2AP, and its fluorescence is measured during folding from secondary structure to tertiary structure / adenine binding.

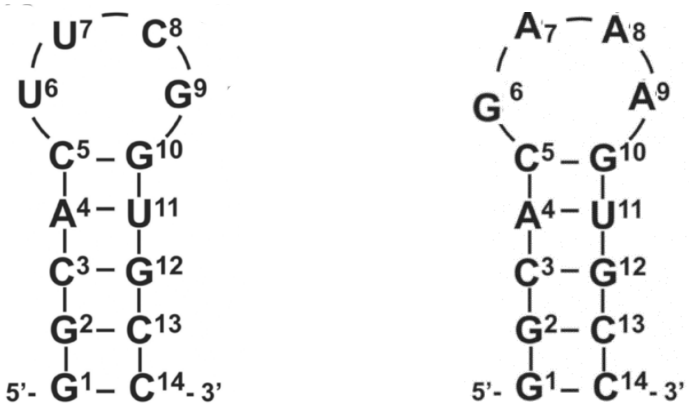


(A) What is the story of this riboswitch structure/function that is revealed by 2AP fluorescence? Explain.

(B) If the order of addition were reversed (*i.e.*, adenine before Mg^{+2}), what might you expect to see and what might it reveal?

Question 8. (10 points; A=6 points, B=4 points)

The thermodynamic stability ($\Delta G°$) of the two RNA tetraloops shown below is very different.



(A) Which structure (left *or* right) is more stable? Explain your answer.

(B) Compare the RNA functions of the two tetraloops.

Question 9. (10 points; A=6 points, B=4 points)

$^{5'}$GCUGCAGAAG$^{3'}$
$^{3'}$CGAUCGCUUC$^{5'}$

The RNA duplex shown above contains a site for binding of the Arginine Rich Motif (ARM) peptide T_{34}RQARRNRRRRWRERQR$_{50}$.

(A) Show where you think the peptide binding site is located. Explain your choice.

(B) What is arginine contributing to RNA binding?

Question 10. (10 points; A=6 points, B & C=2 points)

(A) List three advantages of electron microscopy (cryo-EM) over traditional X-ray crystallography for biomolecular structure determination.

(B) Briefly explain what a Fourier Transform does, and why it is useful in cryo-EM and X-ray crystallography.

(C) Explain what a Constrast Transfer Function (CTF) does as part of the cryo-EM structure determination procedure.