Biology 5357

Chemistry & Physics of Biomolecules

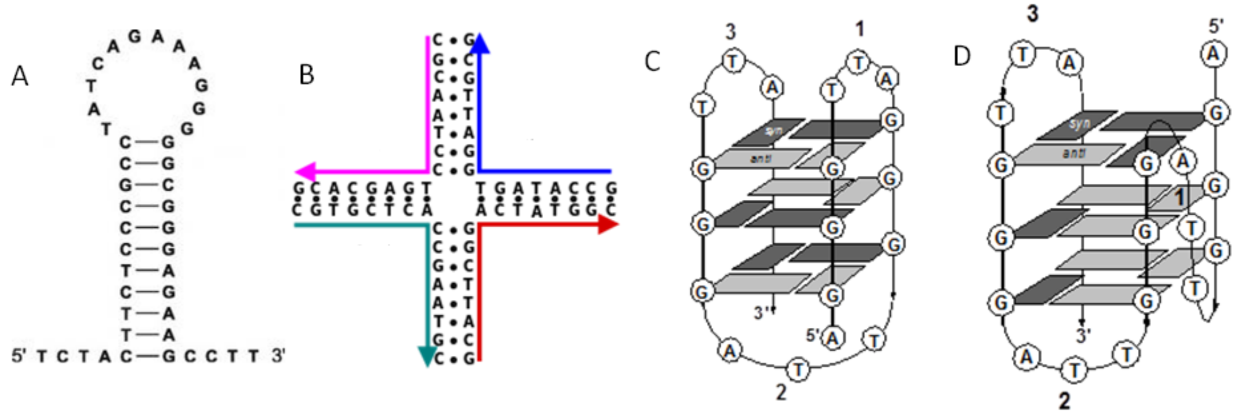
Examination #2

Nucleic Acids Module

November 2, 2018

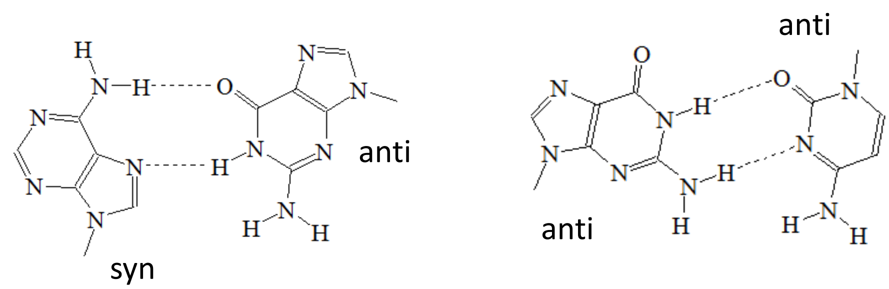
Answer Key

Question 1. (8 points; A-D, 2 pts each)



Hairpin | *Cruciform or Holliday junction* | *Basket G-quadruplex or antiparallel* | *Hybrid G-quadruplex or mixed*

Question 2. (4 points)

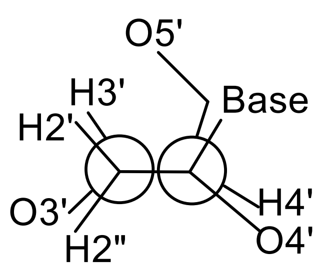


Antiparallel | *Parallel*

Question 3. (6 points; A-B, 3 pts each)

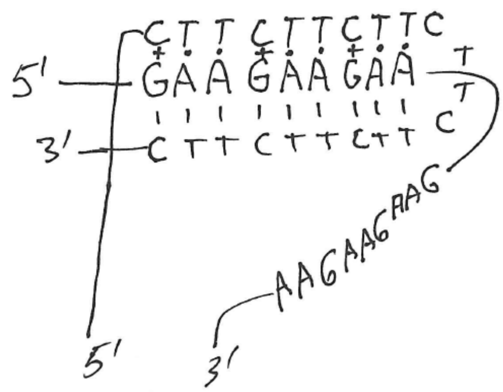
(A) *O4'-exo*

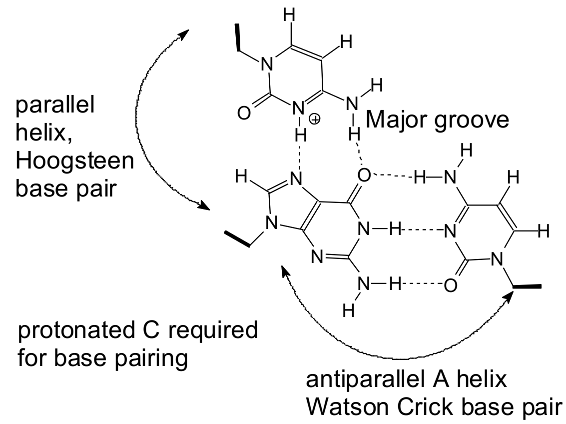
(B) This structure represents the highest energy conformation of a deoxynucleoside. The C2'-C3' bond is eclipsed, and the base and C5' occupy pseudo-axial positions leading to a bad steric interactions between H8 and H5',H5", O5'.



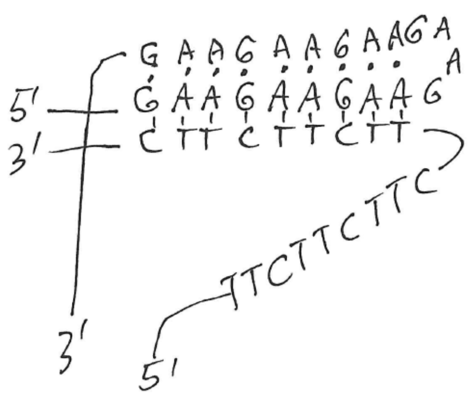
Question 4. (9 points; A-C, 3 pts each)

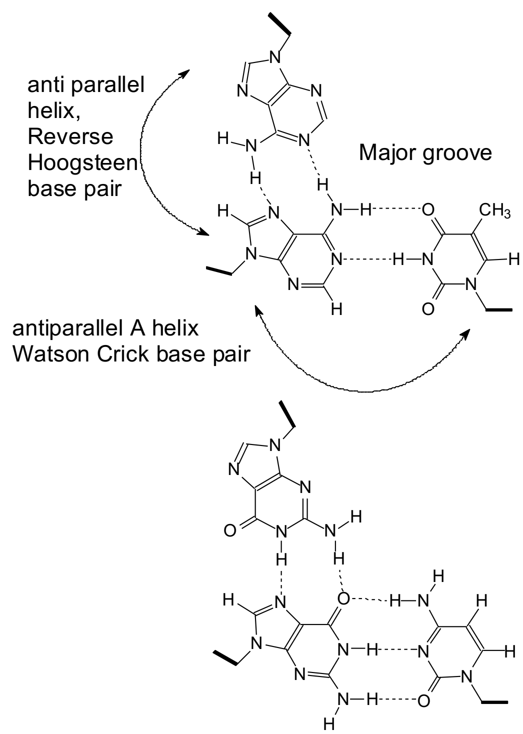
(A)





(B)





(C) *The purines in the purine motif triplex will not be reactive to diethylpyrocarbonate, while the displaced polypyrimidine track will be reactive to* $KMnO_4$ *or* OsO_4*.*

The pyrimidines in the pyrimidine motif triplex will not be reactive to $KMnO_4$ *or* OsO_4*, whereas the displaced polypurine track will be reactive towards diethylpyrocarbonate.*

Question 5. (6 points; A-B, 3 pts each)

(A) *Chain entropy is decreased as the end-to-end distance is increased resulting in an increase in free energy.*

(B) *DNA will get stiffer (i.e. its spring constant will increase). Entropic effects become more important with increasing temperature. Also, in the equation, one can see that as T increases, the end-to-end distance (x) will decrease at a constant force (F) meaning that the effective spring constant (k) must have increased (F = kx).*

Question 6. (4 points)

The stiffness (spring constant) increases with increasing force. DNA has a force-dependent spring constant. This results in a non-linear force-extension plot.

The plot should show a tangent line to the force-extension curve to indicate the stiffness at a particular force.

Question 7. (6 points)

Refraction of light upon interacting with a bead possessing a higher index of refraction than the buffer.

The gradient of laser power in a gaussian beam leads to a net restoring force on the bead towards the center of the beam. This traps the bead in two-dimensions.

The third dimension (along the propagation axis of the laser) is trapped via the offsetting effects of the scattering force pushing the bead downstream (generated by laser reflection) and the gradient force pulling the bead upstream (generated by refraction of a highly focused beam).

Diagrams similar to the ones from the slides shown in class would be acceptable.

Question 8. (7 points; A-G, 1 pt each)

The root-mean-square radius of gyration of an ideal polymer…

(A) is smaller than the root-mean-square end-to-end distance — ☒ True ☐ False

(B) is the radius of the average sphere that comprises each monomer — ☐ True ☒ False

(C) can be measured with SAXS — ☒ True ☐ False

(D) can be measured with FRET — ☐ True ☒ False

(E) is the radius at which the Gaussian distribution of the radii of gyrations adopted by the polymer has maximum probability — ☐ True ☒ False

(F) scales as the root-mean-square end-to-end distance — ☒ True ☐ False

(G) is the radius of the largest sphere that comprises each monomer — ☐ True ☒ False

Question 9. (15 points; A-E, 3 pts each)

(A) $Lc = b * N = 0.63 * 23 = 14.49\ nm$

(B) $\langle R^2 \rangle = 2\ lp\ lc = 2 * 3 * 14.5\ nm = 87\ nm$

(C) $L_K = 2\ lp = 6\ nm$

(D) *A good solvent*

(E) *Twice the mean-squared end-to-end distance of the answer to part B*

Question 10. (4 points)

The Flory χ parameter describes the balance between solvent-solvent, protein-protein, and solvent-protein interactions.

Question 11. (7 points)

Consider the equilibria shown below:

[S1S2] ⇔ [S1] + [S2] + x[M+]   *i.e.*, Duplex ⇔ Single Strands + Salt

Then increasing the salt concentration will move the equilibrium toward duplex via mass action. The value of "x" from counterion condensation theory is 0.18. ***(3 points)***

[P] + [N} ⇔ [P • N] + [M+]   *i.e.*, Protein + Nucleic Acid ⇔ Complex + Salt

The protein has associated salt (anions) and nucleic acid has associated salt (cations) in their uncomplexed states. To form a complex, the ions are replaced by the binding partner (electrostatic binding) to give a net release of salt. So adding salt competes for charge associations and shift the equilibrium to the left, disrupting the complex and decreasing its stability. ***(4 points)***

Question 12. (24 points; A-C, 8 points each)

(A) *The Mg^{+2} concentration affects the docking rate (12X) more than the undocking rate (3X), suggesting that the Mg^{+2} does not increase the stability of the docked state.* ***(2 points)*** *The increase in the rate of docking could be due to more efficient shielding of the charges* ***(3 points)*** *in both stems to allow loser approach that ultimately leads to more efficient docking.* ***(3 points)***

(B) *Yes, there will be a difference in the rates.* ***(2 points)*** *The longer linker would suffer from a more structured Adenosine stack,* ***(3 points)*** *which could impede docking. It will also have an increased entropic penalty for constraining a longer strand, which could show up in a reduced rate of docking.* ***(3 points)*** *Since $\Delta G = -RT \ln(K)$, these rates are related to the overall thermodynamics, so if there is a larger unfavorable entropy, the free energy of the interaction would be less favorable.*

(C) *Yes, there will be a difference in the rates.* ***(2 points)*** *Since U_7 will be more flexible than A_7, the docking rate could increase if the U's are less rigid that the A's and allow different geometries of the approaching tetraloop that still result in productive docking.* ***(3 points)*** *Or, the U's could be so flexible that there is a reduction in the rate of docking since the tetraloop is seldom in the correct orientation for docking. Once docked, the undocked rates should be similar to those for A_7.* ***(3 points)***