

Biology 5357
Chemistry & Physics of Biomolecules
Examination #2

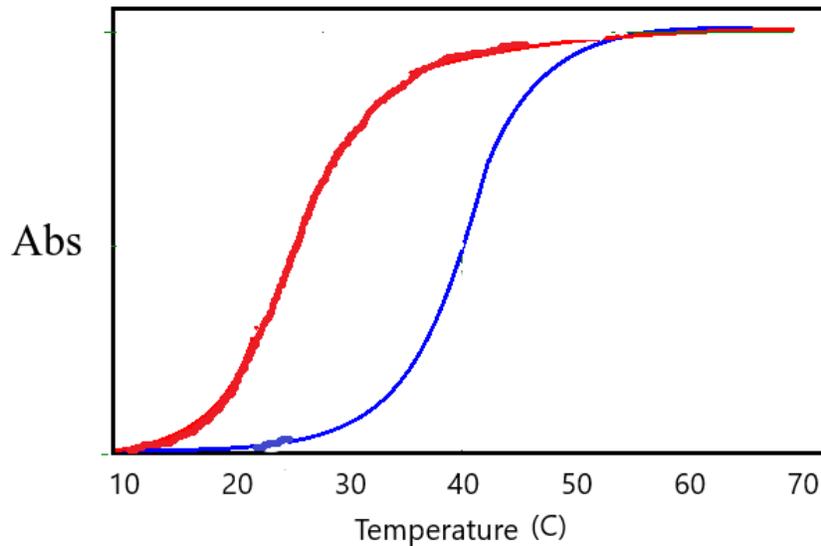
Nucleic Acids Module

November 4, 2022

Name: _____

Question 1 (30 points; A-F = 5 pts each)

The plot below of absorbance vs. temperature shows melting curves for a DNA and an RNA hairpin. Their sequences are the same (except T's or U's), the stem is 7 base pairs, and the loop is 5 nucleobases. The solution conditions are 100 mM KCl, 10 mM HEPES, pH 7.0, with $[NA] = 1 \mu\text{M}$.



(A) At what wavelength would absorbance be measured? Why?

(B) What physical phenomena account for the shape of the curves?

- (C) Label each curve with the correct nucleic acid. Explain your choice.
- (D) Estimate the T_M of each melt.
- (E) If the hairpin concentrations were 10X higher, how would their T_M change? Explain your answer.
- (F) If the KCl concentration were 10X higher, how would the T_M change? Explain your answer.

Question 2 (20 points; A-B = 10 pts each)

RNA transesterification reactions, especially in the presence of Mg^{2+} ions, are the major mechanism of strand scission in an RNA strand.

(A) Illustrate this reaction.

(B) How was it used to probe ligand binding to a riboswitch?

Question 3 (10 points)

A sequence-specific dsDNA binding protein does not bind dsRNA. What is the physico-chemical basis of this discrimination?

Question 4 (10 points)

Ribonucleotides embedded in the genome are the most abundant lesions in living organisms. They are misincorporated by DNA polymerase, and they are removed by RNase H2. RNase H2 is an essential enzyme, and its deletion is embryonic lethal in mice. How would you think a single rN would perturb the DNA?

Question 5 (10 points; A-B = 3 pts each, C = 4 pts)

(A) Fill in the dotted boxes below to provide the missing elements from the 3D Gaussian chain distribution for the end-to-end distance.

$$P_{GC}(R_{ee}) = 4\pi R_{ee}^{\square} \left[\frac{3}{2\pi \langle R_{ee}^{\square} \rangle} \right]^{\square} \cdot \exp \left[-\frac{\square}{\square} \frac{R_{ee}^{\square}}{\langle R_{ee}^{\square} \rangle} \right]$$

- (B)** Which of the following can be described with the same equation?
(for each, circle either YES or NO)

The distribution of internal distances in a freely jointed chain.

YES NO

The distribution of the radius of gyration of the freely jointed chain.

YES NO

The distribution of the end-to-end vector of a freely rotating chain.

YES NO

- (C)** What is the expression of the free energy of an ideal chain?

Question 6 (10 points; A-B = 2 pts each, C-D = 3 pts each)

- (A)** Describe the concept of the scaling exponent.

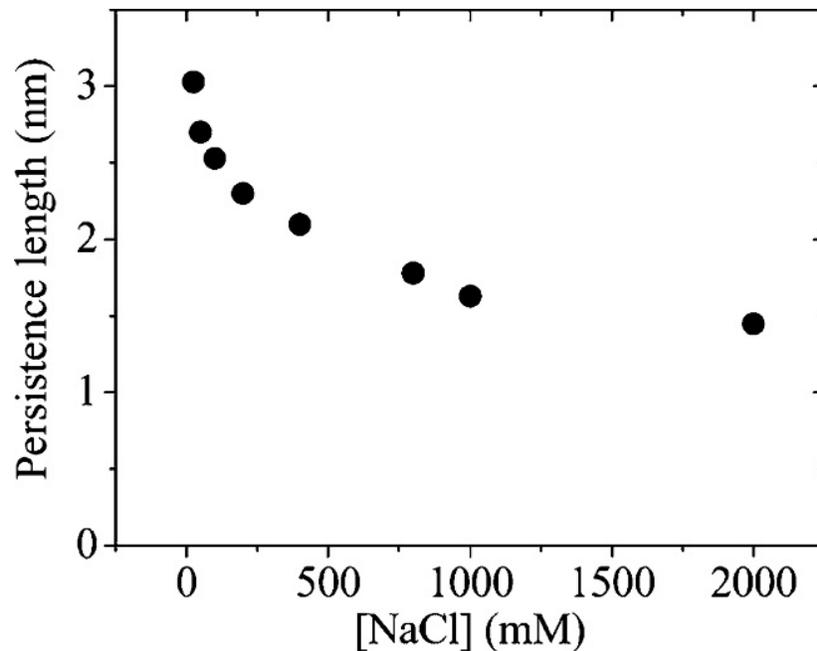
- (B)** Explain the three notable values that can be adopted by the scaling exponent.

(C) Discuss the different solvent regimes and chain behaviors depending on the scaling exponents.

(D) Can you identify a solvent condition that favors the chain expansion of a protein? Explain.

Question 7 (10 points; A = 4 pts, B-C = 3 pts each)

In an article by Murphy, *et al.*, (*Biophysical Journal*, **86**, 2530-2537, 2004) the persistence length of poly-(dT)₂₃ was measured as function of NaCl concentration.



(A) Can you explain why the persistence length is decreasing with increasing salt concentration?

(B) If each nucleotide as a “bond” length of 6.3 Angstroms, what is the mean square end-to-end distance at 0 and 2000 mM NaCl?

(C) What is the corresponding mean square radius of gyration?