

**Biochemistry 661**

**Your Name:** \_\_\_\_\_

**Nucleic Acids, Module I**

**Prof. Jason Kahn**

**Exam I (100 points total)**

**September 25, 2012**

You have 60 minutes for this exam.

Exams written in pencil or erasable ink will not be re-graded under any circumstances.

Explanations should be concise and clear. I have given you more space than you should need. There is a extra space on the last page if you need it.

You do not need a calculator for this exam, and no other study aids or materials are permitted.

Generous partial credit will be given, *i.e.*, if you don't know, guess.

Honor Pledge: At the end of the examination time, please write out the following sentence and sign it, or talk to me about it:

"I pledge on my honor that I have not given or received any unauthorized assistance on this examination."

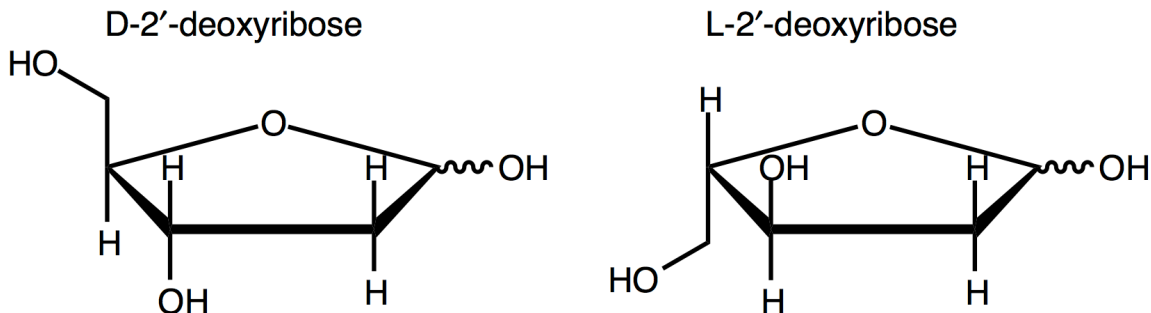
1. (16 pts) You have sequenced an RNA isolated from an extract that helps carry out splicing. You would like to know its secondary structure. Describe how you would proceed. Additionally, you would like to identify base pairing interactions between your new RNA and potential splicing partners. What technique(s) could allow you to catch them? (More space on next page if you need it.)

Score for the page \_\_\_\_\_

2. (6 pts) Why don't sequence-specific RNA binding proteins recognize a fully double-stranded A-form RNA helix?

3. (8 pts) The MAGE paper described a technique that allows rapid genome engineering/evolution. Briefly describe the advantages of this technique as opposed to either traditional cloning or random mutagenesis in generating new or enhanced phenotypes.

4. (18 pts) Here is the structure of L-2'-deoxyribose vs. the usual D-2' -deoxyribose.



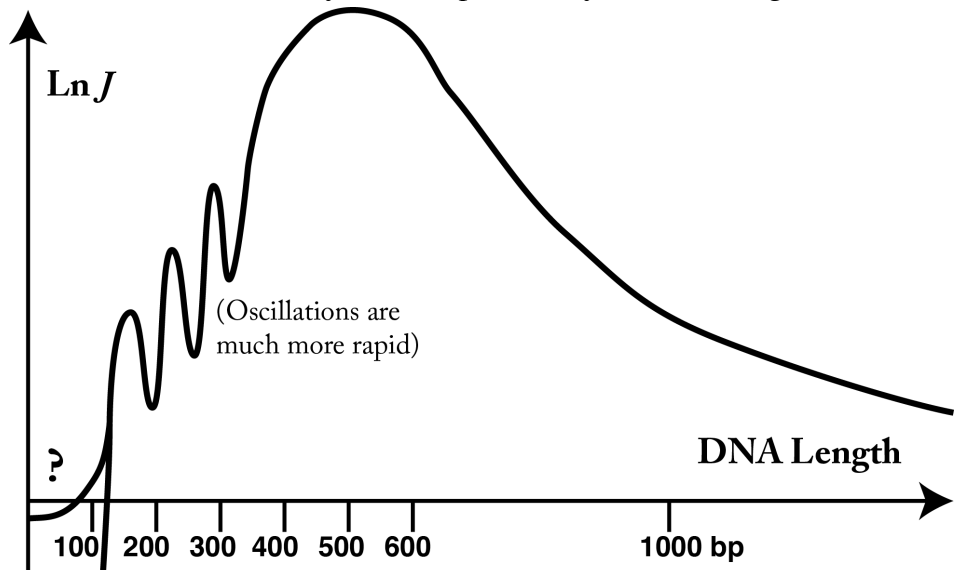
Draw an A-T base pair with L- instead of D-deoxyribose, including both sugars and the four pendant phosphates (three of them just as P's with a circles around them). Sketch the B-form double helix that would result, with strand directions indicated.

5. (7 pts) When we talked about DNA flexibility I said, “There is no ‘the structure’ there.” What does this mean? Define a “persistence length.”

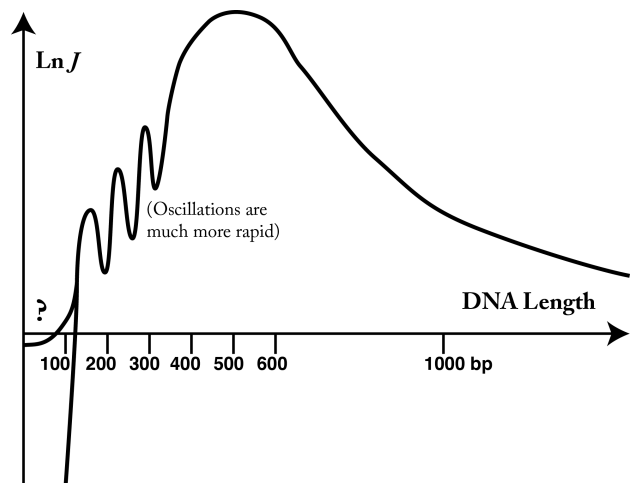
6. (12 pts) The T4 ligase mediated cyclization assay provided a conclusive demonstration that DNA is a double helix in solution. The Shore and Baldwin curve of cyclization probability vs. DNA length is sketched here.

Label the part of the curve that shows rigid rod behavior and the part that looks like random coil.

Sketch on the figure how we can measure the DNA helical repeat from this curve.



On the curve to the right, sketch what the curve would look like if the DNA were significantly stiffer, in terms of torsion only.



7. (15 pts) The portion of the table shown from Borer et al., 1974, provides evidence that a model for hybridization thermodynamics based solely on base composition is inadequate. What is the reasoning for this conclusion? They introduced a nearest-neighbor model for thermodynamics that has subsequently been elaborated by many groups. What is the basic idea of nearest-neighbor thermodynamics? Any scientific theory must be falsifiable (if a theory isn't falsifiable, we call it religion). How could the nearest-neighbor theory have been falsified?

Molecule†	$T_m$ (°C) at‡	
	10 $\mu\text{M}$	100 $\mu\text{M}$
$\text{A}_4\text{CG} + \text{CGU}_4$	-13.9	-1.3
$\text{U}_2\text{CGA}_2$	1.6	11.3
$\text{A}_2\text{CGU}_2$	10.8	22.1
$\text{A}_4\text{G}_2 + \text{C}_2\text{U}_4$	14.0	22.8
$\text{A}_2\text{CGU}_2$	19.6	28.3

8. (18 pts) Negatively supercoiled DNA can extrude quadruplex segments of DNA. Give a sequence that could form a quadruplex. What structure can the other strand form? Sketch a plasmid with and without an extruded quadruplex to explain why negative supercoiling has this effect. Give estimates for before-and-after  $\Delta T_w$  and  $\Delta W_r$ .

Page	Score
1	
2	
3	
4	
5	
6	
Total	

Score for the page \_\_\_\_\_