You have 52 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 extra space on the last page if you need it.
You will need a calculator for this exam. No other study aids or materials are permitted. 
Generous partial credit will be given, i.e., if you don't know, guess.

Useful Equations:

\[ K_a = [H^+][A^-]/[HA] \]  
\[ K_b = [HA][HO^-]/[A^-] \]  
\[ pH = -\log([H^+]) \]  
\[ K_w = [H^+][OH^-] \]  
\[ \Delta G^{\circ} = -RT\ln K_w \]  
\[ R = 0.08206 \text{ L}\cdot\text{atm/mole K} \]  
\[ T = 0 \text{ C} = 273.15 \text{ K} \]  
\[ \ln K_w = -\Delta H^\circ/(RT) + \Delta S^\circ/R \]  
\[ \Delta S - q/T \geq 0 \]  
\[ R = 8.314 \text{ J/mole K} = 1.987 \text{ cal/mole K} \]  
\[ S = k\ln W \]  
\[ \Delta G = \Delta H - T\Delta S \]  
\[ E = \Sigma \epsilon_i n_i \]  
\[ W = N!/(\Pi n_i!) \]  
\[ n/n_0 = \exp[-(\epsilon_f-\epsilon_0)/kT] \]  
\[ N = \Sigma n_i \]  

Chemical standard state: 1 M solutes, pure liquids, 1 atm gases

Biochemical standard state: pH 7, all species in the ionic form found at pH 7

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. **(24 pts) Multiple choice: Circle the best answer for each question**

(a: 4 pts) A configuration in statistical thermodynamics is:

1. A description of the position and velocity of each particle in a system.
2. A set of microstates that all share the same set of observables P, T, n, etc.
3. All of the possible microstates that have a given value of total E.
4. All of the possible microstates that could describe a system at any value of E.

(b: 4 pts) The Boltzmann distribution describes:

1. The distribution of energy among particles in the microstates of the predominant configuration.
2. The distribution of microstates among different ensembles.
3. The distribution of coin flips for small numbers of coins.
4. The distribution of entropy among different microstates.

(c: 4 pts) Boltzmann defined entropy as $S = k_B \ln W$ because

1. It's a postulate - it doesn't need to be justified.
2. Using a logarithm is the only way to make entropy an extensive state function that increases as W increases.
3. The value of “$k_B$” could be chosen to make the numerical values of S fall into an arbitrary and convenient range.
4. All of the above.

(d: 4 pts) Exothermicity per se makes reactions tend to be favorable because

1. It accompanies bond breaking
2. It decreases the total energy of the universe
3. It increases the entropy of the surroundings
4. None of the above

(e: 4 pts) Protein folding from a random coil to a native tertiary structure occurs with

1. Positive $\Delta H^\circ$ and negative $\Delta S^\circ$
2. Positive $\Delta H^\circ$ and positive $\Delta S^\circ$
3. Negative $\Delta H^\circ$ and positive $\Delta S^\circ$
4. Negative $\Delta H^\circ$ and negative $\Delta S^\circ$

(f: 4 pts) The temperature at which $W = 1$ for a system and therefore $S = 0$ is

1. Unknowable
2. Absolute zero
3. 100 °C
4. Infinite

Score for the page__________
2. (30 pts) Short-answer questions

(a; 12 pts) Briefly describe the ergodic hypothesis and why it is hard to test in practice.

- The ergodic hypothesis holds that any microstate that has the same total energy is as likely to occur as any other, so that if we wait long enough all possible microstates will occur. Or - we observe a given configuration at random according to the properties of the total microstates in the configuration.

- It's hard to test because we can’t actually observe microstates and because we may not want to wait the lifetime of the universe for rare ones to show up.

(b; 8 pts) List one biotechnology technique that requires nucleic acid hybridization, and briefly and qualitatively describe why/how nucleic acid thermodynamics can be applied to the design of the oligonucleotides used.

- DNA sequencing
- PCR
- microarrays
- prediction of RNA structure

For example -

We choose PCR sequencing primers according to the T_m desired, or we choose experimental conditions based on the predicted T_m. The sequences can be made longer or shorter or designed to complement a different area according to the thermodynamics of perfect matches vs. mismatches

4 for any one, and there are other correct answers

2 for any change

Score for the page __________
(a: 12 pts) Sketch a van’t Hoff plot for the temperature dependence of the equilibrium constant for an endothermic ordering reaction A+B -> C. If there is a region of the graph corresponding to the reaction being spontaneous, indicate where it is. If there is no such region, explain why not.

\[ \ln K_e = \frac{\Delta G}{RT} \]

- **intercept:** \( \frac{\Delta S^*}{R} < 0 \Rightarrow \text{ordering} \\
- **slope:** \( \frac{-\Delta H^*}{R} < 0 \Rightarrow \Delta H^* > 0 \Rightarrow \text{endothermic} \\
- There is no area where reaction is favored because \( \Delta G > 0 \) for all values of \( T \) - the entropy of the universe can never be increased by this run.

**Instructions:**
1. for idea of a straight line plot
2. for each axis label
3. for \( \odot \) intercept on y-axis
4. for \( \odot \) slope
5. for stating there is no area where reaction is favorable
6. for explanation in terms of \( \oplus \) free energy change or failure to increase \( S_{\text{universe}} \)

Score for the page______ _______
The chart on the left below shows one possible distribution of 48 quanta of energy among 24 molecules, similar to our discussion of microstates in class.

(b; 3 pts) Write down but do not evaluate a numerical expression for the number of microstates \( W \) for the configuration shown on the left above.

\[
W = \frac{N!}{(n_0!)(n_1!)(n_2)!} = \frac{24!}{13! \cdot 2! \cdot 1! \cdot 4! \cdot 4!} = \frac{24 \cdot 23 \cdot 22 \cdot \ldots \cdot 14}{13 \cdot 12 \cdot 11 \cdot 10}\]

(c; 6 pts) Now, consider transferring one quantum of energy from one of the molecules with 7 quanta to one of the ones with 4, as indicated by the arrows in the chart on the left above. This will yield molecules with 5 and 6 quanta. Write down the expression for \( W \) for this new configuration. By what factor did \( W \) increase upon this small redistribution of energy? You do not need to evaluate \( W \) to figure this out.

\[
\frac{W_2}{W_1} = \frac{13! \cdot 2! \cdot 1! \cdot 4! \cdot 4!}{13! \cdot 2! \cdot 1! \cdot 3! \cdot 1! \cdot 3!} = \frac{4 \cdot 4}{1} = 16
\]

Score for the page
(d; 5 pts) If we continue to redistribute energy among the particles, we will reach the equilibrium state described by the Boltzmann distribution. By drawing circles in the chart on the right above, add in a reasonable guess for the energy levels of the 13 remaining molecules according to Boltzmann,

\[
\begin{align*}
1/3 &= 5 + 4 + 2 + 1 + 1 \\
    &= 6 + 2 + 1 + 1 + 1 \\
    &= 4 + 4 + 3 + 1 + 1 \\
    &= 5 + 3 + 2 + 2 + 1
\end{align*}
\]

all are acceptable — any decreasing set of 5 numbers that add up to 13.

(e; 5 pts) Clearly W would be maximized if every \( h_i \) were equal to 1 or 0. Why doesn’t every isolated system just evolve to maximum entropy by placing every molecule on its own unique level? Write down the equation from the front page that expresses this constraint.

- The energy has to come from somewhere — at low \( T \) / low \( E \), there just isn’t enough energy and most of the particles will be in the ground state.

-\( Z \) \( E \) \( n \) = \( E \)

4. (25 pts) Applied thermodynamics: RNA folding

The folding of a single-stranded RNA into a stem-loop is simpler than hybridization of two separate strands to make a duplex. Consider the equilibrium \( UN(\text{folded}) = S(\text{tem}) L(\text{oop}) \) below. For the reaction as written,

\[
\Delta H_f = -52000 \text{ J/mol and} \\
\Delta S^o = -150 \text{ J/mol K}
\]

(made up but reasonable numbers).

(a; 3 pts) Calculate the free energy change \( \Delta G^o \) at 37 °C.

\[
\Delta G^o = \Delta H^o - T\Delta S^o = -52000 + (273.15 + 37)(150) = -3478 \text{ J/mol} \quad \text{(for either)}
\]

\[
=-5800 \text{ J/mol}
\]

(b; 6 pts) Write down the equilibrium constant expression and calculate its value at 37 °C.

\[
K_{eq} = \frac{[SL]}{[UN]} = \frac{-\Delta G^o}{RT} = \frac{-3478}{8.314 \times 290.15} = 8.37
\]

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(c; 6 pts) Calculate the percentage of the RNA in the SL form at 37 °C.

\[
\frac{[\text{SL}]}{[\text{UN}]} = \frac{8.37}{9.37} = 89\% 
\]

(d; 2 pts) The melting temperature \( T_m \) is the temperature at which 50% of the RNA is in the SL form and 50% is in the UN form. What is the equilibrium constant for \( \text{UN} \rightarrow \text{SL} \) at the \( T_m \)? (Note that this is not the same \( K_a \) as for formation of duplex from separate strands, which we treated in lecture.)

\[
K_{eq} = \frac{[\text{SL}]}{[\text{UN}]} = 50 = 1 
\]

(e; 2 pts) What is \( \Delta G^o \) at the \( T_m \)? (Again, differs from the form we saw for duplex formation)

\[
\Delta G^o = -RT_m \ln K = 0
\]

(f; 6 pts) Calculate the \( T_m \). (Again, differs from the form we saw for duplex formation)

\[
\Delta G^o = \Delta H^o - T_m \Delta S^o
\]

\[
T_m = \frac{\Delta H^o}{\Delta S^o} = \frac{-52000 \text{ J/mol}}{-150 \text{ J/mol-K}} = 347 \text{ K} = 73.5 ^\circ \text{C}
\]

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