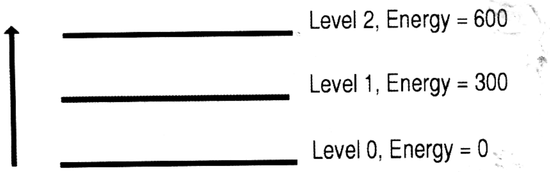
Q1. (30 Points)

(A) (10 points) Consider a molecular system with three energy levels as given below:



The units of energy are such that the Boltzmann constant, $k_B = 1$, and the temperature is 300 K.

There are a total of 1000 molecules in the system. Calculate the number of molecules in each level at equilibrium. Show each step of the calculation.

The probability of being in level j is given by:

$p_j = e^{-E_j/kT} / Q$ where Q (partition function) $= \sum_j e^{-E_j/kT}$

Here, $Q = e^{-0} + e^{-300/300} + e^{-600/300}$

$= 1 + 0.368 + 0.135 = 1.503$

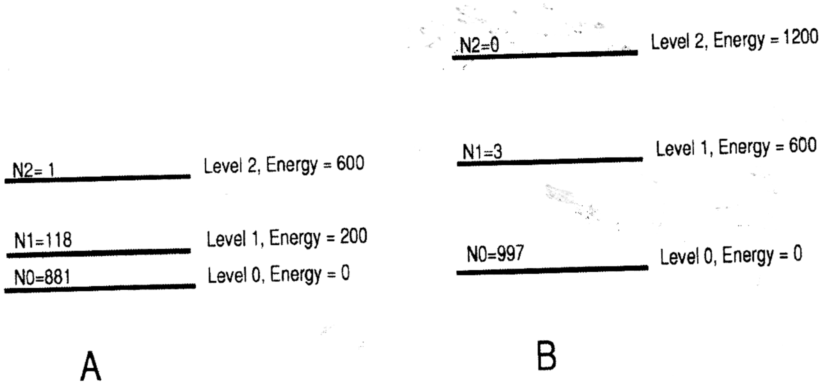
No. of molecules in level 0 = $e^{-0} \times \frac{1}{1.503} \times 1000 = 665$

" 1 = $e^{-1} \times \frac{1}{1.503} \times 1000 \doteq 245$

" 2 = $e^{-2} \times \frac{1}{1.503} \times 1000 = 90$

Question 1, continued.

(B) (10 points) Consider a molecular system A, as shown below, which is converted to a molecular system, B. At equilibrium, at 300 K, the number of atoms in each energy level are shown in the diagram.
Assume that $k_B = 1.0$
Calculate the Helmholtz free energy change for converting A to B. Will the A-> B transformation be spontaneous?



The Helmholtz free energy change, $\Delta A = \Delta E - T\,\Delta S$, where E is the energy, T is the temperature and S is the entropy.

Energy of state A $= 881 \times 0 + 118 \times 200 + 1 \times 600 = 24200$

Energy of state B $= 997 \times 0 + 600 \times 3 + 0 = 1800$

$\Delta E = E(B) - E(A) = -22400$

Entropy: $S = -k_B \sum_j p_j \ln p_j$ where p_j is the probability.

For state A, $p_0 = 0.881$, $p_1 = 0.118$, $p_2 = 0.001$

$S(A) = (-0.881 \ln(0.881)) + (-0.118 \ln(0.118)) + (-0.001 \ln(0.001))$

$= 0.112 + 0.252 + 0.007 = 0.371$ (per molecule)

$S(B) = (-0.997 \ln 0.997) + (0.003 \ln 0.003) = 0.02$ (per molecule)

$\Delta S = [S(B) - S(A)] \times 1000 = -351$

$\Delta A = -22400 + 300 \times 351 = 82900$

$\boxed{\text{NOT SPONTANEOUS}}$

Question 1, continued.
(C) 10 points.

When a protein molecule folds up, the entropy of the protein chain is reduced. Nevertheless, we say that the folding of most protein molecules driven in part by an increase in entropy. Provide a detailed explanation of why this is the case.

Hydrophobic sidechains/groups on the protein reduce the entropy of water when they are exposed in the unfolded protein.

This is because the water molecules cannot hydrogen bond to these groups, and this reduces the rotational freedom of the water molecules

When the protein molecule folds up the hydrophobic groups are buried, and the entropy of the water increases

Q2. (30 points) A protein undergoes a folding/unfolding equilibrium as follows:

$$U(unfolded) \Leftrightarrow F(folded)$$

The equilibrium concentrations of unfolded and folded protein at two different temperatures are given below. The total protein concentration is 2.0 x 10^{-3} M.

Temperature (K)	Concentration of unfolded protein (M)	Concentration of folded protein (M)
300	5.1 x 10^{-6}	2.0 x 10^{-3}
350	2.8 x 10^{-4}	1.7 x 10^{-3}

(A) (10 points) Calculate ΔH° and ΔS° for the folding reaction, assuming that these values are temperature independent. Does entropy favor or oppose folding in this case?

Vant Hoff equation: $\ln \frac{K_2}{K_1} = \frac{\Delta H^0}{R}\left(\frac{T_2 - T_1}{T_1 T_2}\right)$

$T_1 = 300K$, $K_1 = 2.0\times10^{-3}/5.1\times10^{-6} = 0.392\times10^{3}$

$T_2 = 350K$, $K_2 = 1.7\times10^{-3}/2.8\times10^{-4} = 0.607\times10 = 6.07$

$K_2/K_1 = \frac{6.07}{392}$

$= 0.0155$

$\ln K_2/K_1 = -4.168$

$\Delta H^0 = \ln(K_2/K_1) R\left(\frac{T_1 T_2}{T_2 - T_1}\right)$

$= (-4.168)(8.314)\left(\frac{300 \times 350}{50}\right) = -72\,710.8\ J = -72.8\ kJ/mole$

$\Delta G^0 = -RT\ln K$ at 300K, $\Delta G^0 = -8.314 \times 300 \times \ln(0.392\times10^{3})$

$= -14.89\ kJ/mole$

$\Delta G^0 = \Delta H^0 - T\Delta S^0 \Rightarrow \Delta S^0 = -\left(\frac{\Delta G^0 - \Delta H^0}{T}\right) = -0.193$

Entropy opposses the reaction

Question 2, continued.

(B)(10 points) The unfolding temperature of the protein is defined as the temperature at which the protein is half unfolded. What is the unfolding temperature of this protein?

When the protein is half unfolded the free energy change for the reaction is zero.

$$\Delta G^0 = 0 = \Delta H^0 - T\Delta S^0$$

$$\Rightarrow T = \frac{\Delta H^0}{\Delta S^0} = \frac{-72.8}{-0.193} = 377K$$

Question 2, continued.

(C) 10 points.

Consider an enzyme that speeds up the rate of U (unfolded) -> F (folded) by a factor of 2.0 at 300 K. What would be the effect of this enzyme on the rate of the reverse reaction: F(folded) -> U(unfolded)? Explain your answer.

The enzyme, as a catalyst, does not affect the equilibrium constant for the reaction.

Since $K = \frac{k(\text{folding})}{k(\text{unfolding})}$, if the folding rate is increased by 2.0, the unfolding rate must also increase by a factor of 2.0

Q3. (30 points)
Consider the simple Michaelis-Menten mechanism for an enzyme catalyzed reaction:

$$E + S \underset{k_{-1}}{\overset{k_1}{\leftrightarrow}} ES \xrightarrow{k_2} E + P$$

k_1 and k_{-1} are very fast.
$k_2 = 100\ s^{-1}$, $K_M = 1.0 \times 10^{-4}$ at 280 K
$k_2 = 200\ s^{-1}$, $K_M = 1.5 \times 10^{-4}$ at 300 K.
K_M, the Michaelis constant, is given by: $K_M = \frac{k_{-1} + k_2}{k_1}$

(A) (10 points) For [S] = 0.1 M and $[E]_{tot} = 1.0 \times 10^{-5}$ M, where $[E]_{tot}$ is the total enzyme concentration, calculate the rate of formation of product at 280 K.

Since the substrate concentration is so high we can assume that the enzyme is saturated, so

$$[ES] \approx [E]_{TOT}$$

and $\nu = [E]_{TOT}\, k_2 = 100\ \text{sec}^{-1} \times 1.0 \times 10^{-5}\ \text{M}$
$= 1.0 \times 10^{-3}\ \text{M sec}^{-1}$

(B) (10 points) What is the value of the activation energy for k_2?

Use the Arhenius equation:

$$\ln \frac{k_2}{k_1} = -\frac{E_a}{R}\left(\frac{1}{T_2} - \frac{1}{T_1}\right)$$

$$\Rightarrow \quad E_a = -R \ln\left(\frac{k_2}{k_1}\right)\left(\frac{1}{T_2} - \frac{1}{T_1}\right)^{-1}$$

$$= -8.314 \ln\left(\frac{200}{100}\right)\left(\frac{1}{300} - \frac{1}{280}\right)^{-1}$$

$$= -8.314 \times 0.693 \times (-4200)$$

$$= 24198\ \text{J/M} = +24.198\ \text{kJ/mole}$$

Question 3, continued.

(C) (10 points) Under conditions of steady state, when the concentration of ES is constant, derive a relationship between the initial velocity or rate of the reaction, v, and the Michaelis constant, K_M.

$$E + S \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} ES \xrightarrow{k_2} E + P$$

$$\frac{d[ES]}{dt} = k_1 [E][S] - k_{-1} [ES] - k_2 [ES]$$

$$= k_1 [E][S] - (k_{-1} + k_2)[ES]$$

Under steady state conditions,

$$\frac{d[ES]}{dt} = 0 \Rightarrow k_1 [E][S] = k_{-1} + k_2 [ES]$$

$$\Rightarrow [ES] = \frac{k_1}{(k_{-1}+k_2)} [E][S]$$

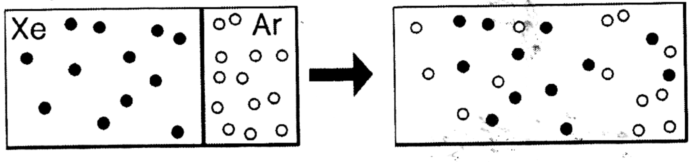
$$[ES] = \frac{[E][S]}{K_M}$$

The rate of the reaction

$$v = k_2 [ES] = \frac{k_2}{K_M} [E][S]$$

(Further rearrangements yield the normal form of the Michaelis-Menten equation, but this is not necessary)

Q4. (25 points)
Consider a fixed volume container that initially contains 3.0 moles of xenon at 0.50 atm and 300K separated from 4.0 moles of argon at 3.0 atm and 300 K. The barrier separating the gases is removed, while maintaining constant temperature (illustrated below).



(A) (10 points) Calculate the change in the entropy in going the intial to final conditions that arises due to the change in pressure. Assume ideal gas behavior, and show all steps of the calculation, and the units of your final answer.

The change in entropy due to the change in pressure is $\Delta S = nR\ln\left(\frac{V_2}{V_1}\right)$

For Xe, initial volume $V_1(Xe) = \frac{n_{Xe}RT}{P_1(Xe)} = \frac{3.0 \times 0.08206 \times 300}{0.5} = 147.7$ L

For Ar, initial volume $V_1(Ar) = \frac{n_{Ar}RT}{P_1(Ar)} = \frac{4.0 \times 0.08206 \times 300}{3.0} = 32.8$ L

Total volume = 180.5 L = $V_2(Ar) = V_2(Xe)$

For Xe, $\Delta S = 3.0 \cdot 8.314 \cdot \ln\left[\frac{180.5}{147.7}\right] = 5.0$ J/K

For Ar, $\Delta S = 4.0 \cdot 8.314 \cdot \ln\left[\frac{180.5}{32.8}\right] = 56.7$ J/K

$\Delta S_{TOT} = 61.7$ J/K

Question 4. Continued.

(B) (10 points) What fraction of the total change in entropy is due to the entropy of mixing?

$$\Delta S_{mix} = -R(n_{Xe}\ln(X_{Xe}) + n_{Ar}\ln(X_{Ar}))$$

$$\boxed{X_{Xe} = \frac{3.0}{3.0+4.0} = 0.43} \quad \boxed{X_{Ar} = \frac{4.0}{3.0+4.0} = 0.57}$$

$$\Delta S_{mix} = -8.314\,(3\ln 0.43 + 4\ln 0.57) = 39.74\ \mathrm{J/K}$$

Fraction due to mixing: $\frac{39.74}{39.74+61.7} = .39$

(C) (5 points) As ideal gases, the argon and xenon atoms do not attract each other. Consider a situation where the argon atoms attract each other slightly, and the xenon atoms attract each other slightly, but the argon and xenon atoms do not attract each other. Would the entropy of mixing be smaller or greater for such a situation than your answer in (B)? Explain your answer.

Qualitatively, we would expect less mixing in this situation, and so the entropy change will be smaller. Xenon atoms will be more likely to be around other Xenon atoms, likewise for Argon atoms.

Q5. (25 points)

Consider a membrane-bound pump that couples ATP hydrolysis to the secretion of H^+ out of the cell, operating under conditions where the concentration of H^+ outside the cell is 0.18 M, the internal pH is 7 and the temperature is 300 K.
(A) (15 points) What is the change in free energy for the transport of 1 M of H^+ from inside the cell to outside, under these conditions, when the membrane potential is 0.06 V (inside negative)?

Work done against the potential = nFV,
where F is the Faraday constant

$= +1.0 \times 96500 \times 0.06$

$= 5790$ J/mol

Work done against the concentration gradient:

$$= RT \ln\left(\frac{[H^+]_{out}}{[H^+]_{in}}\right) = 300 \cdot 8.314 \cdot \ln\left[10^{7} \cdot 0.18\right]$$

$= 35{,}924$ J/mol

$$\Delta G_{TOT} = (41{,}714 \text{ J/mole}) \times (1 \text{ mole})$$

$= 41{,}714$ J

Question 5, continued.

(B) (10 points) Consider the following redox reactions:

$NAD^+ + H^+ + 2e^- \rightarrow NADH$ ($E^{o\prime} = -0.32$ V)

$1/2\ O_2 + 2H^+ + 2e^- \rightarrow H_2O$ ($E^{o\prime} = 0.816$ V)

If we could couple the free energy released by these reactions to the proton pump, how much NADH (in moles) would have to be oxidized by oxygen to transport one mole of H^+ across the membrane?

For the reaction,

$NAD^+ + H^+ + 2e^\ominus \rightarrow NADH$

Free energy change = $\Delta G^o = 2 \times F \times -0.32$

$= 61760$ J/mol

For the reverse rxn,

$\Delta G^o = -61,76$ kJ/mol

For the 2nd rxn,

$\Delta G^o = 2 \times 96500 \times 0.816$

$= -157$ kJ/mol

Total ΔG^o : $-219, 248$ J/mol

$\frac{41,714}{219,248} = 0.1902$ moles of NADH

would have to be oxidized.

Q6. (35 points) Consider a chemical reaction in which the reactant, A, reacts to form product, P. A plot of the reciprocal of the concentration of A versus time is a straight line. When the initial concentration of A is 1.0 x 10^{-2} M, its half-life is found to be 30 minutes.

(A) (10 points) What is the order of the reaction? Explain the reasoning behind your answer.

$$\frac{1}{[A]} \propto t \quad \text{or} \quad [A] = \beta t^{-1}$$

where β is a constant.

$$t^{-1} = [A]/\beta$$

$$\Rightarrow \frac{d[A]}{dt} = -\beta t^{-2} = -\beta\left[\frac{[A]}{\beta}\right]^{2}$$

$$\text{Rate} = -\frac{d[A]}{dt} = +\frac{1}{\beta}[A]^{2}$$

Since the rate depends on the square of the concentration, the reaction is second order.

(B) (5 points) Write a one line reaction scheme that is consistent with the kinetics.

$$A + A \rightleftharpoons A\cdots A \xrightarrow{fast} 2P$$

or $$A + A \rightleftharpoons A\cdot\cdot A \xrightarrow{fast} A + P$$

(C) (10 points) What is the value of the rate constant for your mechanism in part (B)? What is the unit of the rate constant

The rate law for a 2nd order reaction is $\frac{1}{[A]} = kt + \frac{1}{[A]_0}$ When $[A]_0 = 1\times10^{-2}$ M, $t_{1/2} = 30$ minutes

$$\Rightarrow \frac{1}{[A_0]/2} = k\,30 + \frac{1}{[A_0]}$$

$$\Rightarrow 2 = k\,30[A_0] + 1$$

$$\Rightarrow k = \frac{1}{30[A_0]} = \frac{1}{30\ 1\times10^{-2}} = \frac{100}{30} = 3.33\ \text{mole}^{-1}\ \text{min}^{-1}$$

Question 6, continued.
(D) (10 points)
When the initial concentration of A is 5.0 x 10^{-3} M, what will be the half life of the reaction?

Now solve for the time when $[A] = \frac{1}{2}[A_0]$

$$\frac{2}{[A_0]} = kt + \frac{1}{[A_0]}$$

$$\Rightarrow \quad 2 = kt[A_0] + 1$$

$$\Rightarrow \quad kt = \frac{1}{[A_0]} \qquad t_{\frac{1}{2}} = \frac{1}{[A_0]k} = \frac{1}{5.0\times10^{-3} \times 3.33}$$

$= 60.06$ minutes

Q7. 25 points

Consider a binding equilibrium between a drug, D, and a receptor, R:

$D + R \Leftrightarrow DR$

When the concentration of the drug is 10^{-9} M, the receptor is 33% saturated with the drug.

(A) 10 points. Assuming that each receptor molecule binds one drug molecule, what is the dissociation constant for the binding reaction?

$$\text{Fractional Saturation} = \frac{[D]}{K_D + [D]} = 0.33$$

$$\Rightarrow [D] = 0.33 K_D + 0.33 [D]$$

$$\Rightarrow K_D = \frac{0.66 [D]}{0.33} = 2 [D] = 2 \times 10^{-9}$$

(B) 5 points. What are the units of the dissociation constant?

The equilibrium constant is dimensionless. It has no units because we always implicitly divide all the concentrations by the concentration of the standard state.

$$K_D = \frac{[D][R]}{[DR]} \text{ is actually } \frac{\frac{[D]\text{moles}}{1\text{ mole}} \frac{[R]\text{moles}}{1\text{ mole}}}{\frac{[DR]\text{moles}}{1\text{ moles}}}$$

Question 7. Continued.
(C) 10 points. By how much should the binding energy of the drug be increased at 300 K so that the saturation of the receptor is at least 99% at the same concentration of the drug?

In terms of Y, the fractional occupancy or saturation,

$$Y = \frac{[D]}{K_D + [D]}, \quad 0.99 = \frac{[D]}{K_D + [D]}$$

$$0.99\,K_D + 0.99[D] = [D]$$

$$K_D = \frac{0.01[D]}{0.99} = 0.01[D] = 10^{-11}$$

Initially, $\Delta G^\circ = -RT\ln \underset{\nearrow(2\cdot 10^{-9})}{K_D} = 49.959$ kJ/mol

Finally, $\Delta G^\circ = -RT\ln[10^{-11}] = 63.16$ kJ/mol

Additional Binding energy required =

$63.16 - 49.959 = 13$ kJ/mol

-END OF EXAM-