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Chemistry 130A - Midterm 2

50 minute open book exam

November 8, 1993

Read the whole test, then do the easiest parts first.

1. (15 points)

(a) Radioactive tritium, T = $^3H_1$, has a half-life of 12.5 years (6.57 X 10^6 minutes, 4,550 days). How many days will it take for the number of disintegrations per minute (dpm) to decrease by 1%?

(b) How many dpm will take place in 1 gram of pure T_2O?

(c) T_2O can be used to measure the volume of water in the circulation of an animal. One mL of saline solution with some T_2O in it was injected into a vein of the animal. The dpm of the injected solution was 1.37 X 10^6. After 5 minutes some blood was removed and the dpm of 1 mL of water from the blood was found to have 2.10 X 10^2 dpm. Calculate the volume of water (in L) in the circulation of the animal.

2. (10 points)

The stoichiometry for a reaction is: A→ D. A possible mechanism is:

$$A \underset{k_2}{\overset{k_1}{\rightleftarrows}} B \quad \text{fast to equilibrium}$$

$$B + C \xrightarrow{k_3} D + C \quad \text{slow}$$

(a) Write a differential equation for the formation of intermediate B.

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2. (b) Because both k_1 and k_2 are very large compared to k_3 , the ratio of concentrations of B to A can be set equal to an equilibrium constant K. Write a differential equation for the formation of product D which depends only on [A], [C], k_3, K, and time. What is the order of the kinetics with respect to [A], with respect to [B], with respect to [C]?

3. (25 points)
(a) The vapor pressure of water for a saturated NaCl solution (1 Kg NaCl in 1 L water) is 15 torr. What is the water vapor pressure when 1 Kg more of solid NaCl is added to this solution?

(b) If we change the NaCl solution in problem (a) to a saturated solution of camphor, which is less soluble in water than NaCl, will the water vapor pressure increase, decrease, or not change from the answer to problem (a)? Explain.

(c) Some amino acids in a protein are changed by site-directed mutagenesis. The isoelectric points of the protein P and its mutant M are pH 6 and pH 11, respectively. Their SDS gel electrophoresis reveals a similar molecular weight of about 80,000. At pH 7 which molecule (P or M) has the higher positive charge? Name one type of amino acid whose removal would explain the difference betwee M and P.

(d) The ratio of the sedimentation coefficients s_P/s_M for protein P and mutant M in problem (c) is 0.950, and the ratio of the diffusion coefficients, D_P/D_M, is 0.955. All measurements are done in the same solvent at the same temperature. Assuming that the specific volumes of P and M are the same, calculate the ratio of their molecular weights, M_P/M_M.

(e) Besides sedimentation, diffusion and electrophoresis what other method can we apply to measure the absolute molecular weight for protein P and its mutant M?

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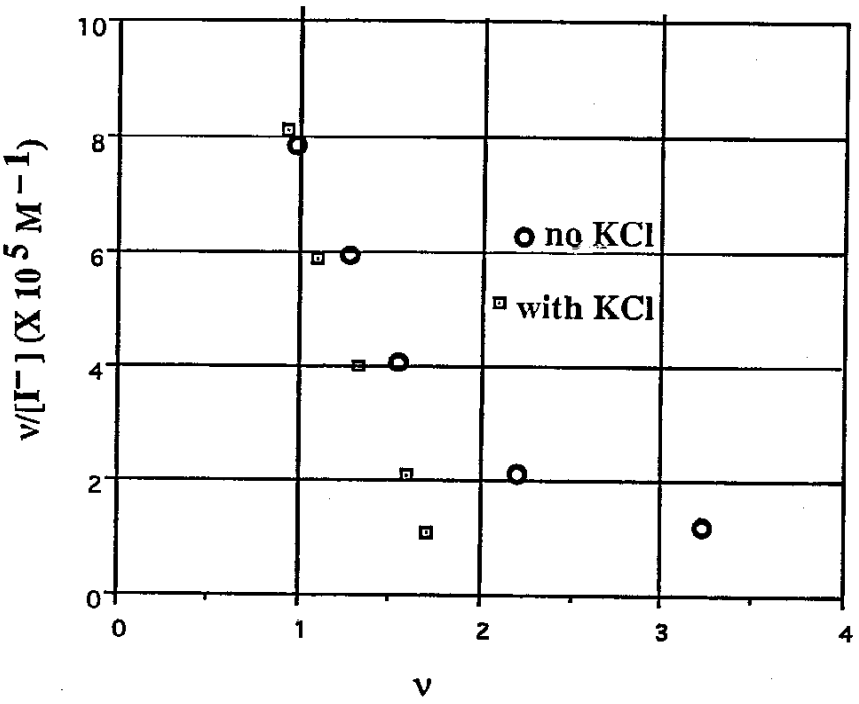
4. (25 points)

A protein P was thought to play a role in the metabolic uptake of iodide ion, therefore it was important to measure its ability to bind iodide. An equilibrium dialysis experiment was done in which the purified protein was placed on one side of a membrane and allowed to reach equilibrium with different concentrations of iodide ion. In the first series of experiments only the pure protein and KI were present. The data O were plotted as the Scatchard plot shown below (labeled no KCl). A second series of experiments was done in which 1 M KCl was also present. The data ▫ are also shown below (labeled with KCl).



(a) In one experiment (not plotted above) the following data were measured.

Total concentration of protein inside dialysis membrane = 1.00 μM.

Total concentration of I^- outside dialysis membrane = 0.43 μM.

Total concentration of I^- inside dialysis membrane = 0.95 μM.

Calculate ν, the number of I^- bound per protein molecule, and $\nu/[I^-]$, the number bound divided by the free I^- concentration.

(b) From the Scatchard plot done in the presence of 1 M KCl calculate the number of binding sites for I^- and the equilibrium constant (with units) for binding. You may assume that the sites are independent and identical.

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4. (c) From the Scatchard plot in the absence of KCl, what can you conclude about the number of binding sites and their equilibrium constants? Are there more or less sites than in the presence of KCl? Are the binding constants stronger, weaker or the same?

(d) Why should KCl have any effect if it is not involved directly in the binding of I^-.

5. (25 points)
(a) Native gel electrophoresis can be used to measure the equilibrium constant for binding a protein molecule to a DNA.

$$DNA + P \overset{K}{\rightleftharpoons} Complex$$

Describe how you would do the experiment. What solutions are applied to the gel? What is measured? How is this related to the equilibrium constant? What is the crucial assumption made in this experiment?

(b) Define in words what osmotic pressure is.

(c) What is the osmotic pressure in atm of a 0.15 M solution of NaCl at 37 °C?

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5. (d) A difference in pH across a membrane can cause a potential difference to be produced. What is the potential difference in volts (measured by a hydrogen electrode) produced at equilibrium by a concentration of 0.050 M HCl inside a cell and a concentration outside of 0.093 M HCl? The temperature is 37 °C. Which side is more positive?

(e) If there is no voltage difference, what is the difference in chemical potential in kJ for the HCl inside and outside the cell?. The temperature is 37 °C. Which solution has the higher chemical potential?