Thermodynamics, pH and Buffers Homework Chem 331 Biochemistry

Name _____ (_____ / 80 points)

For each question show all calculations and simply describe the process. Use complete sentences, bullets or outlines will NOT be accepted. For formula weights, pKa, Ka or other information needed for the problem, start with your book then look for other sources.

1) **(5 points)** Calculate the pH of a 1 L solution containing 100 mM formic acid and 125 mM sodium formate before and after adding 1ml of 5M NaOH. How much would the pH have changed if the NaOH were added to 1 liter of pure water?

2) **(5 points)** What is a Good's Buffer? (don't forget, I google and wiki too!) Be specific and look for detail in your explanation.

3) **(5 points)** You need a buffer at pH 7.5 for use in purifying a protein at 4°C. Using Tris, pK 8.08, you carefully make up 0.01 M Tris buffer, pH 7.5 at 25°C and store it in the cold to equilibrate it to the temperature of the purification. When you measure the pH of the buffer at 4°C, you see the buffer has increased to pH 8.1. What is the explanation for this increase? How can you avoid the problem?

4) **(5 points)** Phosphate buffered saline (PBS) is a commonly used buffer. Sometimes with calcium and magnesium, other times without. Describe a PBS buffer and explain the function of each component in the buffer. Calculate how to make a 1 L solution of PBS. Describe the steps involved including which apparatus and glassware you would use and when. See question 35 for a guide to this part of the question.

5) **(2.5 points)** Draw the structure **and** net charge of Histadine for the predominant molecular species at **each** of the following pHs 2, 7, and 10

6) **(7.5 points)** The pKa's of amino acid functional groups can change depending on microenvironment. If the side chain of glutamate were in a hydrophobic pocket, what would you predict the shift in pKa to be and why? How would a pKa of an acidic amino acid change if the carboxyl group we very close to a basic amino acid?

7) An enzyme-catalyzed reaction was carried out in 250 ml of a 10 mM Hepes buffer, pH 7.90. As a result of the reaction, 0.5 mole/liter of H^+ was consumed.

a) **(2.5 points)** What were the concentrations of each form of the buffer at the start of the reaction?
b) **(5 points)** What was the pH at the end of the reaction?
c) **(2.5 points)** Was this an appropriate buffer for this experiment? Why?

8) **(5 points)** Draw the reaction for the bicarbonate buffer system. Describe what happens to the equilibria of this reaction when a person has kidney disease which slows or even stops filtering. Write the clinical description of what this person will be in and then describe how the patient will compensate. Be specific in terms of biochemistry.

9) **(10 points)** Consider the following interconversion, which occurs in glycolysis:

Fructose-6-phosphate ←→ Glucose-6-phosphate K'eq = 1.97

a. What is the $\Delta G°'$ for the reaction at 25°C?
b. If the concentration of fructose-6-phosphate is adjusted to 1.5 M and the concentration of glucose-6-phosphate is adjusted to 0.5M what is the ΔG?
c. Why are $\Delta G°'$ and ΔG different (hint: list the similarities/differences)?

10) **(10 points)** The synthesis of the activated form of acetate (acetyl-CoA) is carried out in an ATP-dependent process:

Acetate + CoA + ATP ---> acetyl-CoA + AMP + PP_i

a. The $\Delta G°'$ for the hydrolysis of acetyl-CoA to acetate and CoA is -32.2 kJ/mol and the $\Delta G°'$ of hydrolysis of ATP to AMP and PPi is -30.5 kJ/mol. Calculate the $\Delta G°'$ for the ATP-dependent synthesis of acetyl-CoA.
b. Almost all cells contain the enzyme inorganic pyrophosphatase, which catalyzes the hydrolysis of PP_i to P_i. What effect does the presence of this enzyme have on the synthesis of acetyl-CoA?

11) **(5 points)** Phosphoglucomutase catalyzes the reaction in which a phosphate group is transferred from the C-1 of glucose to the C-6 of glucose (G1P ⇆ G6P). A student incubates a 0.2 M solution of glucose-1-phosphate overnight with a small amount of the enzyme. At equilibrium the concentration of glucose-1-phosphate is 9.0×10^{-3} M and the concentration of glucose-6-phosphate is 19.1×10^{-2} M.

Calculate the equilibrium constant (K_{eq}) and the standard state free energy ($\Delta G°'$) for this reaction at 25°C.

12) **(10 points)** Using information from class and the linked article from Scientific American, examine and explain the statement, "*we hum along smoothly, beneficiaries of a 3 kcal/mol energy window*…".