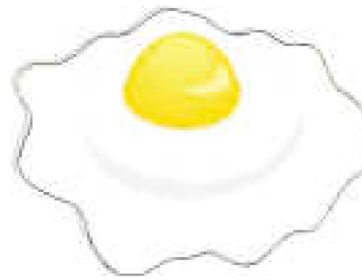
#3: Buffers and Titrations

Why are buffers important in our bodies and in nature?

By this point, you are familiar with the concepts of acids and bases, and that of pH. Changes in pH—even small ones—can have these results, among others:



- Change the structure of molecules and macromolecules—for example, disrupting hydrogen bonds and other intermolecular forces that give proteins their 3-dimensional shape. It's possible to "cook" an egg simply by adding acid to it. (You're actually denaturing the proteins in the egg, making them lose their 3-D shape, unraveling, and getting tangled in one another as evidenced by the egg whites going from clear to white. Normally, you'd use heat to denature your egg.)

- Affect the solubility of salts and minerals. Acid rain changes the soil pH; nutrients on which plants depend become more soluble and leach from the soil. Aluminosilicates dissolve, releasing aluminum ions, which are toxic to fish. Limestone (calcium carbonate) statues slowly dissolve.

Part I: What do you know about acids, bases, and buffers in the "real world"?

Read the introduction, which provides a few examples of how changes in pH can affect molecular structure and can cause minerals to become more soluble (which can impact living organisms). Where have you encountered buffers? Write down some examples of "real world" buffer systems you know about that function to resist changes in pH. You may have studied such systems in biology and geology courses . . .

Part II: Regulating blood pH

The pH of blood plasma needs to remain within a very narrow range. Normal blood pH is 7.40. Death can result if the pH of your blood goes below 7.0 or above 7.8. The most important equilibria that affect (and regulate) the pH of blood plasma are:

$$CO_2\,(g) + 2\,H_2O\,(l) \rightleftharpoons H_2CO_3\,(aq) + H_2O\,(l) \rightleftharpoons H_3O^+(aq) + HCO_3^-(aq) \qquad (1)$$

Take a minute to think through what it means to have two chemical equilibria linked to each other. How does a change in CO_2 concentration affect H_3O^+ concentration, for instance? In nature, it is very common to have several chemical equilibria occurring simultaneously so that a change at one end affects something else several steps down the line.

You'll often see these equilibria simplified as: $CO_2\,(g) + H_2O\,(l) \rightleftharpoons H_3O^+(aq) + HCO_3^-(aq)$ (2) because carbonic acid is unstable in water and immediately decomposes back to CO_2 and H_2O. Biochemists tend to just think of this equilibrium as occurring between CO_2 and HCO_3^- as in the second equation.

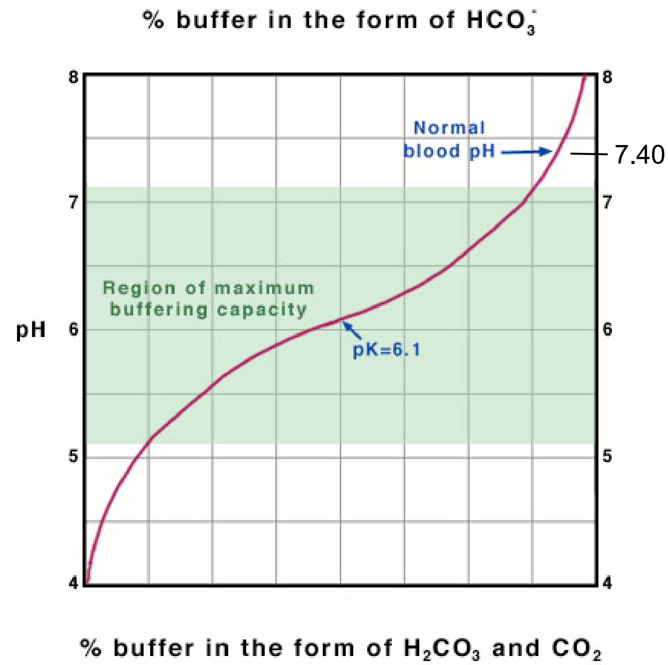
Your blood pH can change (hopefully only slightly) due mainly to stresses that affect the equilibrium system described above. Fortunately, this system provides a buffer, and your body has other mechanisms in place to deal with situations in which too much acid or too much base enters the bloodstream.

1. Read each description below. Predict how ***that activity alone*** (without a counter mechanism) would affect your blood's pH. Explain your reasoning.

Description	Potential change in blood pH (circle one):	Clearly explain your reasoning. It may be helpful to use both words and chemical equations.
1a. Your cells convert glucose to produce energy through aerobic respiration: $C_{12}H_{12}O_6 + O_2 \rightarrow CO_2 + H_2O$ + energy	pH increases pH stays the same pH decreases	
1b. During rigorous exercise, you begin to produce lactic acid.	pH increases pH stays the same pH decreases	
1c. During rigorous exercise, you begin to exhale more vigorously.	pH increases pH stays the same pH decreases	

How are 1b. and 1c. related?????

2. Consider this **portion** of a titration curve for the carbonic acid / bicarbonate buffer system.



Source for graph: http://www.chemistry.wustl.edu/~edudev/LabTutorials/Buffer/Buffer.html

2a. Circle the point on the graph where the buffer consists of 50% HCO_3^- and 50% H_2CO_3/CO_2.

Explain your reasoning.

2b. What does the normal blood pH indicate about the relative ratio of HCO_3^- to H_2CO_3/CO_2 in the blood? Explain your reasoning.

Support your reasoning by performing a calculation.

2c. You overhear a student say, "Since normal blood pH is above 7.00, it must have a higher ratio of base to acid. If it were below pH 7.00, it would have a higher ratio of acid to base." How would you respond?

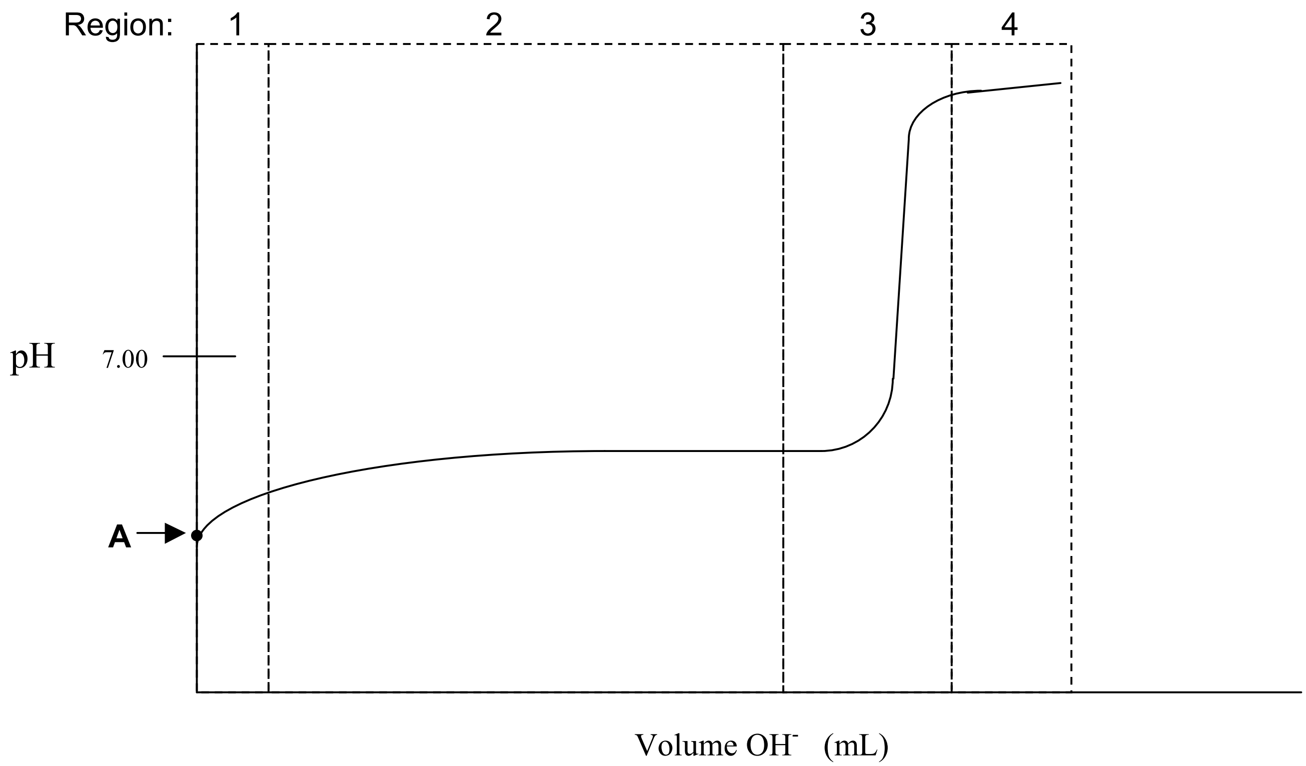
2d. Biochemical experiments often use a buffer system based on tris-(hydroxymethyl)aminomethane ($(HOCH_2)_3CNH_2$), also called "TRIS" or "THAM."

TRIS is a weak base. What is the chemical formula of its conjugate acid?

The pKa of its conjugate acid is 8.075. What mole ratio of acid to base is required to prepare a buffer at the same pH as human blood?

Part III: Titrations and titration curves

The titration curve below is the result of adding strong base (such as NaOH) to a weak acid.



1. Write a chemical equation that represents the weak acid solution at point A, before the titration begins:

   If you knew the Ka and initial concentration of the weak acid, how would you calculate the pH at point A? Describe and/or show the setup:

2. Where is the equivalence point? Mark this point as “B” on the titration curve.
   Explain in words what the equivalence point is.

3. Write a chemical equation that represents the reaction taking place during the titration:

4. Consider these species: HA, A^-, and OH^-
   Identify the major species in solution within each region marked on the graph. It may help to follow the curve, starting a “0.00 mL NaOH” and visualizing what’s going on in solution as the titration progresses.

   Region 1: Region 2: Region 3: Region 4:

5. How would you calculate the pH for any point within Region 2? Explain and/or show the setup.

6. How would you calculate the pH for any point past point B? Explain and/or show the setup.

7. Suppose you don't know the identity of this acid.
   How would you determine its identity from the titration curve?
   Explain in words and by labeling important points on the titration curve.

8. Point B (the equivalence point) should be above pH 7. Why does this make sense?
   Write a chemical reaction to help explain.

Part IV: Additional Practice

Consider the titration of 25.00 mL of 0.1000 M acetic acid with 0.1000 M NaOH.

1. Write a chemical equation for the titration reaction.
2. Determine the volume of NaOH required to reach the end point.
3. Identify the major species in solution and calculate the pH after:

   a. 0.00 mL NaOH added b. 5.00 mL NaOH added c. 12.50 mL NaOH added

   d. 20.00 mL NaOH added e. 25.00 mL NaOH added f. 30.00 mL NaOH added