pH and BUFFERS
Class set- Please return

INTRODUCTION:

The fluids of the body are solutions which consist of the solvent water and dissolved solutes. Hydrogen ions are also present in the body fluids and their concentration can be measured by taking the pH of the solution. pH represents the negative logarithm of the hydrogen ion concentration with a low pH representing a high concentration of hydrogen ions and vice-versa.

An **acid** is a chemical that **donates hydrogen ions** and a **base** is a chemical that **accepts hydrogen ions**. An acid-base reaction always involves a *conjugate acid-base pair* that is made up of the hydrogen ion donor and hydrogen ion acceptor. For example, lactic acid ($CH_3CHOHCOOH$) and lactate ($CH_3CHOHCOO^-$) form the acid and base respectively of a conjugate pair. Together they act as a buffer system. See Figure 1-1.

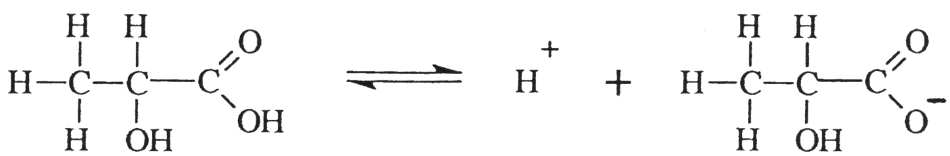

Figure 1-1. Lactic Acid and Lactate Conjugate Pair

The body fluids require a stable pH otherwise proteins will become denatured and nonfunctional. The body fluids achieve pH stability with the presence of buffers. The major buffer of the intracellular fluid in the body uses phosphate as a conjugate pair while the major extracellular buffer is the carbonate pair. These buffers plus others can stabilize the pH by adding or removing hydrogen ions to or from the body fluids as needed.

The addition of hydrogen ions to phosphate ($PO4^{-3}$) from hydrochloric acid utilizes three consecutive conjugate pairs as phosphate is hydrogenated to phosphoric acid (H_3PO4). See Figure 1-2.

Basic Pair	$HPO_4^{-2} \Leftrightarrow H^+ + PO_4^{-3}$
Midrange Pair	$H_2PO_4^{-1} \Leftrightarrow H^+ + HPO_4^{-2}$
Acidic Pair	$H_3PO_4 \Leftrightarrow H^+ + H_2PO_4^{-1}$

Figure 1-2. The Conjugate Pairs of Phosphate.

PROCEDURE:
*Standardize pH meters first!!

Titration of Phosphate and Carbonate:

Obtain 50 ml of 0.01M **Phosphate (Na_3PO4)** and pour it into a 250 ml beaker. Immerse the tip of the clean glass electrode into the solution and measure the pH. Record the pH in the data table. Add 5.0 ml of 0.01M HCl from the buret into the beaker of phosphate. Swirl gently, or use the stir plates, and take the pH of the solution. Continue adding 5.0 ml aliquots of HCl and taking the pH at each volume until **100 ml of** HCl has been added.
Record all data in the **Data Table I.**

Repeat the procedure using 50 ml of 0.01M **Carbonate (Na_2CO3)** in place of the phosphate.

Testing the Buffering Abilities of Various Solutions:

Obtain 20 ml of pond water and pour it into a 100 ml beaker. Take the initial pH and then again after 2.0 ml aliquots of HCl have been added to the pond water. Continue until a total of **20 ml of HCl** has been added to the pond water. Repeat the process using the following solutions in place of the pond water. 20 ml of a 10% glucose solution, 20 ml of a 1% protein solution, 20 ml of distilled water, and 20 ml of artificial blood. Record all data in **Data Table II.**

PROCESSING THE DATA: (Generate two graphs)
1. Plot the data collected from data table I with pH on one axis and milliliters of HCL on the other axis.
2. Plot the data collected from data table II with pH on one axis and milliliters of HCL on the other axis.

CONCLUSION QUESTIONS: Complete sentences please!

1A. What are the *most effective buffering ranges* on graph #1 for phosphate and carbonate?

(*for 1A, 1B, and 1C show analysis lines drawn on graph for credit)

1B. What is the pH at the *midpoint* for both phosphate and carbonate?

1C. At what pH for phosphate and carbonate is the *maximum buffering capacity*?

2. What is the *chemical equation* for the carbonate buffer system in bodily fluids?

3. Based on graph #2, list in order of <u>best to worst</u> the buffering abilities of the five solutions by observing their initial slopes.

4. Based on the graph, discuss the difference between the glucose solution, and the protein solution's ability to buffer. Chemically, why is one a better buffer than the other?

5. What chemicals in the blood make it such an excellent buffer? (see your text for help).

6. What is respiratory acidosis? How does the carbonic acid/bicarbonate buffer system compensate to maintain blood pH?