Respiration and pH (lab 8.4)

a) Background information on respiration and pH

The background information for understanding today's exercise is found in lab 8.4 in your lab manual. Although we will use a different procedure than the one described in lab 8.4, you should read lab 8.4 for the background information.

b) The effect of CO_2 on blood pH

In this activity, you will investigate what effect the CO_2 from your cells has on the pH your blood. The CO_2 in your breath will represent the CO_2 that cells make and a beaker of pink water that you breathe into will represent the blood.

What to do:

1) Get a 250 ml beaker and add 30 ml of water from the flask on the front desk. Label the beaker "Blood with CO_2"
2) Add 1 drop of phenolphthalein pH indicator to the water in the beaker.
3) Add 36 drops of 1M NaOH to the water in the beaker. The solution in the beaker should turn pink. Mix the solution by swirling gently.
4) Use pH paper to take the pH of the solution. Do this as follows:
   a) Tear a square of pH paper.
   b) Use forceps to submerge a square of pH paper into the water for ten seconds. After ten seconds, lift the pH paper out. Hold the wet paper in the air for 30 seconds to allow the color to develop.
   c) After 30 seconds, compare the square to the color key on the pH paper container. Use the color on the "downhill" end of the pH paper.
   d) Record the pH: _____ .
5) Get a straw and vigorously blow bubbles through the water. Keep blowing until all pink color is completely gone. (This may take a 10 minutes. It helps to hold the beaker over white paper to make sure all the pink is completely gone).
6) Use pH paper to take the pH. Record the pH: _____
7) Set aside the "blood" beaker (you will use it in the next activity).
8) Show your instructor your results before continuing.

What does it mean?

The pink water in the beaker represents your blood. You represent a cell that is expelling CO_2 into the blood. (All of your cells expel CO_2 into the blood. That's how cells get rid of the CO_2 they make).

You should have observed that the CO_2 from the cell (you!) lowered the pH of the "blood" in the beaker. In other words, CO_2 acts like an acid when it mixes with blood (or any other liquid that contains water). Recall that acids are molecules that add H^+ ions to a solution. **In the space below, write the balanced chemical equation that shows how CO_2 adds H^+ to the blood.** Hint: The CO_2 reacts with a water molecule.

c) The buffering effect of CO_2 on acids in the blood

In the previous activity, you showed that CO_2 acts like an acid (it decreases the pH of the blood). But CO_2 also has another effect on the blood: It buffers the blood. A buffer is any molecule that stabilizes the pH of a liquid. In other words, when you add a buffer to a liquid, the liquid resists pH change.

In this activity, you will investigate the buffering effect of CO_2 on your blood. The water that you blew bubbles through represents your blood. Keep your first beaker of "blood" (from part b) but get a second beaker of pure water to represent an unbuffered liquid.

What to do:

1) Keep your first beaker of "blood" but get a second 250 ml beaker and add 30 ml of water from the flask on the front desk. Label it "Unbuffered"
2) Use pH paper to take the pH of the Unbuffered water.
   Record the pH: _____
3) Use pH paper to take the pH of the "blood" beaker.
   Record the pH: _____
4) Add 3 drops of 1M HCl (hydrochloric acid) to both beakers. Mix by swirling gently.
5) Use pH paper to take the pH of the Unbuffered water.
   Record the pH: _____
6) Use pH paper to take the pH of the "blood" beaker.
   Record the pH: _____
7) Do the following calculations:

   How many units of pH did the "blood" change when you added the acid? _____

   How many units of pH did the unbuffered water change when you added the acid? _____

8) Show your instructor your results before continuing.

What does it mean?

The water in the "blood" beaker represents your blood. Just like your blood, it has lots of CO_2 dissolved in it (from when you blew bubbles through it in activity B). The water in the unbuffered beaker represents what your blood would be like *if there was no CO_2 in it*.

You added equal amounts of HCl acid to each beaker. Acids decrease the pH of solutions, so you should have observed that the pH of both solutions decreased. However, you should also have observed that the "blood" resisted the acid more than the unbuffered solution. In other words, the blood had much less of a pH change. This is because the CO_2 creates

molecules that buffer the blood from acids (by absorbing the H^+ released by the HCl acid).

In the space below, write the balanced chemical equation that shows how CO_2 buffers the blood from acids by absorbing H^+. Hint: It is not the CO_2 itself that absorbs H^+, but a molecule that CO_2 creates in the blood.

d) The buffering effect of CO_2 on bases in the blood

A base is a molecule that removes H^+ ions from a solution. Therefore, if you add a base to a solution, the pH of the solution will increase.

In the previous activity, you showed that CO_2 buffers the blood from acids. But does CO_2 also buffer the blood from bases? The answer is yes. CO_2 creates a molecule that buffers the blood from bases. The buffering molecule creates H^+ to replace any H^+ that the base absorbed.

In the pre-lab lecture, your instructor gave a balanced chemical equation that showed how that molecule (that comes from CO_2) buffers the blood from bases. **In the space below, write that balanced chemical equation.** Hints: The buffering molecule is not CO_2 itself, but instead a molecule that CO_2 creates in the blood. This buffering molecule buffers against bases by creating H^+.

e) The buffering effect of albumin on the blood

Activities A-D were designed to demonstrate that the CO_2 that enters the blood buffers the blood from acids and bases. However, blood carries many substances in addition to CO_2. One of the most abundant substances in the blood a protein called albumin. Does albumin also play a role in buffering the blood? You will investigate this question.

What to do:

1) Dispose of the solutions in your beakers by pouring them down the drain. Thoroughly wash out all of your beakers with water.
2) Add 30 ml of unbuffered water from the flask on the front desk to the beaker labeled "Unbuffered". Set it aside for now. You will use it in step 8.
3) Add 60 ml of unbuffered water from the flask on the front desk to the beaker labeled "Blood with CO_2". Then add 1 drop of phenolphthalein pH indicator and 6 drops of 1M NaOH (the beaker solution should turn pink) to this beaker. Mix by swirling gently.
4) Get a straw and vigorously blow bubbles through the water in the "Blood" beaker. Keep blowing until all pink color is completely gone. (This may take a few minutes. It helps to hold the beaker over white paper to make sure all the pink is completely gone).

5) Get a clean dry 250 ml beaker. Label it "Blood with CO_2 and albumin". Add one level scoop full of albumin protein to this beaker.
6) Pour 30 ml from the "Blood with CO_2" beaker into the "Blood with CO_2 and albumin" beaker. Stir the beaker with the albumin protein using the straw until the protein completely dissolves.
7) You should now have three beakers: The "Unbuffered" beaker contains only water (no buffers). The "Blood with CO_2" beaker contains water with CO_2 (from your bubbles). The water in the "Blood with CO_2 and albumin" beaker contains CO_2 and albumin. In terms of the molecules it contains, which beaker is most like real blood? _____
8) Use pH paper to take the pH of all three solutions:
   Unbuffered = _____
   Blood with CO_2 = _____
   Blood with CO_2 and albumin = _____
9) Add 3 drops of 1M HCl to each of the three beakers. Mix by swirling gently.

10) Use pH paper to take the pH of all three solutions:
   Unbuffered = _____
   Blood with CO_2 = _____
   Blood with CO_2 and albumin = _____

11) Do the following calculations:
   How many units of pH did the unbuffered water change when you added the acid? _____

   How many units of pH did the blood with CO_2 change when you added the acid? _____

   How many units of pH did the blood with CO_2 and albumin change when you added the acid? _____

12) Show your instructor your results before continuing.

<u>What does it mean?</u>

In step 9 you added equal amounts of HCl acid to each beaker. Acids decrease the pH of solutions, so you should have observed that the pH of all solutions decreased. However, the amount that the pH changed was different for each of the solutions. The solution that changed pH the least must have been the best buffered of the three solutions.

Which solution was buffered the best? _____ Based on your results, does albumin play a role in buffering your blood? Does CO_2 play a role in buffering your blood?