

Laboratory Experiment: Buffers

ver March 18, 2021

Before class

1. What is the definition of a buffer solution; that is, what components must be present in a solution for it to be called a buffer?
2. What is the physical meaning of the word buffer in this context; that is, what behavior should a buffer exhibit? (Do you know what the geopolitical term “buffer state” means?) Describe what should happen to a buffer if you add a few drops of strong acid. And what if you add strong base?
3. **How do the components of the solution give rise to the expected pH behavior of a buffer?** That is, explain your model of buffer behavior. (Chemical, not geopolitical!) Write balanced reactions involving acetic acid or acetate ion to support the explanation. Check your explanation against the explanation in D. C. Harris’s Quant textbook: review the section labeled “effect of adding acid to a buffer.” (Any edition of Harris will do.)
4. Draw a structure for the abbreviation “OAc”. If you didn’t use it before, rewrite your balanced reactions for Question 3 using this abbreviation.
5. Estimate the pH of an acetate buffer.

Experimental investigation of buffers

Part 1: What is buffer behavior?

You have two solutions: an acetate buffer that is 0.5 M HOAc and 0.5 M NaOAc, and a 1M NaCl solution. You will add drops of acid or base to these solutions and compare how the pH changes as a result.

Using plastic droppers, move samples of the solutions to a plastic well plate. You want to fill each well about half full with either the acetate buffer or the salt solution, and you want to have three samples of each solution. Then add a drop of indicator to each solution, either bromocresol green or universal indicator. Now you’re ready for the test: Add a drop of hydrochloric acid to one sample of acetate buffer, and a drop of NaOH to another. Compare the colors of the three samples: Did the acetate buffer change color, and by how much? Now repeat the test on the NaCl solution. How much does the salt solution change color when it is treated with acid or base?

Calibrate a pH probe (Hanna Checker Plus); do a two-point calibration using pH 7 and pH 4 solutions. Press and hold the black button until CAL is displayed. The probe will recognize which calibration solution you are using. Once you have calibrated the probe, measure the pH of your solution samples in the well plate. The probe is small enough that you can use it directly in the well, as long as you didn’t put too much solution in the well at the start. Gently stir the solution with the pH probe as you watch the reading stabilize. Record the stable reading. If you see the reading reverse direction, then it has stabilized.

Write a sentence describing “buffer behavior”?

Part 2: How can you make an acetate buffer?

1. Check the calibration of your pH probe; recalibrate if necessary. As a group, measure the pH of the following solutions: 0.1 M HCl, 0.2 M HOAc, 0.2 M NaOAc, 0.1 M NaOH.
2. As a group, mix together **equal volumes** of solutions in pairs; for example, mix 10 mls of 0.1 M HCl and 10 mls of 0.1 M NaOAc. 10 mL grad cylinders will work fine. There are six unique pairs to measure. Save the mixtures!
3. Fill in the following table with your group's data:

	pH unmixed	w/ 0.2 M HOAc	w/ 0.2 M NaOAc	w/ 0.1 M NaOH
0.1 M HCl				
0.2 M HOAc		-----		
0.2 M NaOAc			-----	
0.1 M NaOH				-----

Analysis

1. As a group, decide which pairs of solutions (the mixtures) formed acetate buffers when they were made.
2. Propose experiments that would confirm that the solutions are, or are not, buffers. As a class, test two mixtures--one that you think is a buffer and one that you think isn't.
3. As a group, write balanced reactions explaining how the buffer solutions were formed after mixing.
4. Based on results in the table, predict what would happen if equal volumes of all four solutions were mixed!
5. To the mixtures you saved, add either one drop of the indicator bromocresol green or three drops of universal indicator. Correlate your observations of the color with the pH values you measured.

Part 3

Testing the Henderson-Hasselbalch equation

In part 1 you mixed solutions of acetic acid and sodium acetate to make an acetate buffer. You used equal volumes of both solutions. Now you will mix together different volumes and look at the effect on pH of the final buffer solution. The pH of a buffer solution is predicted by the Henderson-Hasselbalch equation.

$$pH = pK_a + \log \frac{[A^-]}{[HA]}$$

In the H-H equation, HA is the weak acid and A⁻ is its conjugate base.

Preliminary work

In your journal, sketch a rough graph of a plot of pH vs log([Base]/[Acid]). What values do you predict for the slope and intercept of the graph?

Rewrite the H-H equation as it would apply to an acetate buffer.

Your calculations for the experiment will be made simpler because the solutions of acetic acid and sodium acetate are the same concentration. Rewrite the equation so that the log term contains the key measured quantities that you need to predict the pH of the buffer solutions.

Experimental work

Recalibrate your pH probe.

Your instructor will assign your group three or four different mixtures to make, using 1-10 mls of 0.2 M HOAc and 1-10 mls of 0.2 M NaOAc. Measure the volumes using a grad cylinder, mix the two solutions together and measure the pH of the resulting buffer. After you measure the pH, add a drop of bromocresol green to the solution and record the color. Save the solutions so that you can compare the colors; take a photo if you wish!

Create a graph of your data in Excel. Fit the data to a straight line. Pool the data from all the groups and fit the entire data set to a straight line.

Part 4

When preparing buffer solutions, does it matter how we measure the volumes?

We will now explore the **reproducibility** of making different buffers. We're going to make the buffer solutions by measuring out the acid and base solutions two different ways: Once with graduated cylinders, and again with micropipettors. The goal is to try to make the same buffer solution with the same pH both ways.

Preliminary work

You'll use the 0.2 M HOAc and 0.2 M NaOAc solutions you used in Part 2, and some additional solutions involving phosphate and ammonia. You need to know the pKa's of these chemicals.

Target pH values: 4.35, 5.15, 6.90, 7.50, 8.84, 9.64.

Your group will be assigned two or three target pH values. Your goal is to mix the appropriate acid and base solutions and hit the target pH values. For each of the pH values you are assigned, you will prepare one solution using grad cylinders and another using micropipettes. You'll have to calculate how much of each solution you will need. Check your calculated volumes with the instructor.

Use the **Henderson-Hasselbalch equation** (or **buffer equation**) to figure out how much of each solution to mix.

$$pH = pK_a + \log \frac{[A^-]}{[HA]}$$

Experimental work

You should check the calibration of your pH probe before you begin.

Grad cylinders: Your goal is to mix together enough of each solution so that you hit the target pH AND the final volume of your buffer solution is between 15 and 25 mL. **Mix the calculated volumes of the two solutions together, then measure the pH. Do not add any additional acid or base to the solution once you begin measuring the pH.** Gently stir the solution with the pH probe as you watch the reading stabilize. Record the stable reading.

Micropipettes: There are six micropipettes available which you must share. If you've never used one before, you should practice pipetting water. Your goal is to mix the buffer solution so that you hit the target pH AND the final volume of your solution is between 3 and 10 mL. **Pipette the calculated volumes of the two solutions into the same vessel, then measure the pH. Do not add any more acid or base solution once you begin measuring the pH.** Gently stir the solution with the pH probe as you watch the reading stabilize. Record the stable reading.

Analysis

Pool the data obtained by the class as paired values: (grad cylinder, micropipette). *There might be a significant difference in the pH values you obtain using these two methods.* You will perform a **Paired Sample t test** to find out. In Excel this is a type 1 test using the TTEST function. Your report should include a table with the target pH value, the volumes you mixed, and pHs that you measured, but you should perform the paired sample t test only on the **measured values of pH**. Do not do any statistical tests using the original target values.