How-To-Do-It

# Making pH Tangible

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The concept of pH is an important one for our students. All of our experiments depend upon pH, and nearly every process in living cells depends upon the maintenance of a proper pH level, as enzymes will generally only function within a narrow pH range. Since the synthesis of adenosine triphosphate in living cells depends upon the creation and maintenance of a pH gradient, it is worth noting that nearly all of the energy used by all living systems on this planet comes from pH gradients.

Many students have a difficult time understanding the concept of pH, as it is somewhat abstract, and mathematical on a logarithmic scale. We present here a simple set of experiments to give students practical, hands-on experience with pH. The labs also touch on the mathematics by demonstrating the logarithmic nature of the scale. These experiments are appropriate for high school biology or introductory college-level biology labs.

pH is the measure of the hydrogen ion ($H^+$) concentration of an aqueous solution. In our experiments, as well as within our bodies, we keep the concentration of $H^+$ ions constant by using buffers, chemicals that will compensate for changes in this ion by absorbing excess ions as the pH goes down, or freeing them as the pH goes up. For instance, our blood must maintain a pH level of 7.35–7.45. A slight change can cause severe illness and even death. Therefore blood must have a system with which to control the level of pH. One such system is a bicarbonate buffer system, which consists of carbonic acid, a weak acid, and sodium bicarbonate, its complementary weak base.

$$H_2CO_3 \Leftrightarrow H^+ + HCO_3^-$$

When the concentration of $H^+$ ions increases, they are removed with the formation of carbonic acid. When the concentration of $H^+$ ions decreases, they are replaced by the dissociation of carbonic acid molecules. Therefore, the concentration of $H^+$ ions remains constant, maintaining this range of pH[1].

In this lab, students will test the pH of different substances: acids at various concentrations to demonstrate the logarithmic nature of the scale, and common household substances. They will also study the effect of a buffer on acidic solutions by comparing the behavior of buffered and unbuffered solutions upon the addition of acid. Finally, they will compare common over-the-counter antacid remedies.

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## *Procedure*

### 1. pH of Different Concentrations of Acid

If time allows, students should prepare the following concentrations of acid from 0.1 M HCl stock.

0.01 M HCl
1.0 mM HCl
10 micromolar HCl
1.0 micromolar HCl
Distilled water

Instruct the students to calculate the expected pH of each of these solutions, using the formula **pH = −log[H⁺]** by plugging the concentration of acid into the equation. Remind the students that in addition to the HCl, the water in aqueous solutions contributes $10^{-7}$M $H^+$. For example, for 10 mM HCl:

**10 mM = $10^{-2}$M.** Thus,
**pH = $-\log[10^{-2}] = 2$.**

Use narrow range pH paper to determine the pH of each solution. Record. There will be some variation from the expected pH of each solution, due largely to the inaccuracy of the pH paper. However, all results should be within 1 pH unit of calculated expectations.

### 2. pH of Foods & Household Substances

Have the students predict the pH of each of the following substances, and then test with pH paper:

Vinegar
Lemon juice
White grape juice
Cola (clear, preferably)
Tap water
Distilled water
Milk (diluted with 2 parts distilled water to decrease any interference from the white color)
1% Drano®

For example, the sour taste of vinegar is attributed to acetic acid; thus, vinegar may be expected to have a pH of below 7.

Each student should bring in one additional household substance for testing.

### 3. Buffers

This procedure tests how distilled water, NaCl, and a sodium phosphate buffer solution react to the addition of HCl. Sodium phosphate is chosen because it is a fairly strong buffer and is commonly used in biological experiments. It is also simple to explain its buffering capacity, as the phosphate ion may exist in solution with zero, one, two, or even three $H^+$ ions attached:

$$[PO_4]^{3-} + H^+ \leftrightarrow H[PO_4]^{2-}$$
$$H[PO_4]^{2-} + H^+ \leftrightarrow H_2[PO_4]^-$$
$$H_2[PO_4]^- + H^+ \leftrightarrow H_3[PO_4]$$

The pH indicator Brom Cresol Purple is used to detect a pH change. This indicator should initially be purple and will change to yellow at or below pH 5. You may substitute chlorophenol red, or methyl purple if you prefer.

Table 1. Solutions and reagents.

**To prepare 0.1 M phosphate buffer solution:**
Prepare 0.1 M $Na_2HPO_4$, by dissolving 14.2 g anhydrous $Na_2HPO_4$ in a liter of distilled water.
Prepare 0.1 M $NaH_2PO_4$ by dissolving 12.0 g anhydrous $NaH_2PO_4$ in a liter of distilled water.
Mix 3.5 parts 0.1 M $Na_2HPO_4$ with 1 part 0.1 M $NaH_2PO_4$.
Use dilute NaOH or dilute HCl [such as 10 mM] to adjust the pH to 7.0.
**To prepare 0.1 M NaCl:**
Dissolve 5.8 g NaCl in a liter of distilled water.
The solution should be pH 7, but for this lab, the pH should be checked. If it is more than 0.1 units off, use dilute NaOH or dilute HCl [such as 10 mM] to adjust the pH to 7.0.
**To prepare 0.1 M HCl:**
Most commercial preparations of concentrated HCl are 12 M. Thus, to prepare a 0.1 M solution, add 8.3 ml concentrated HCl to 991.7 ml concentrated HCl.
Note: Distilled water used in the buffers section should also be brought to pH 7.0 using dilute NaOH or dilute HCl [such as 10 mM] before use.
Brom Cresol Purple may be purchased from Fisher Chemicals, catalog #LC11890-7.

Prepare 0.1 M sodium phosphate solution, 0.1 M NaCl, and 0.1 M HCl as described in Table 1.

Each student or group should obtain three test tubes, labeled for 0.1 M sodium phosphate buffer, distilled water, and 0.1 M NaCl. 5 mL of the appropriate solution should then be dispensed into each tube.

Add 3–4 drops of indicator dye to each tube and mix until homogeneous.

Test each tube by adding 0.1 M HCl dropwise, mixing the solution after each drop. Continue to add acid until the solution changes color, indicating that the solution has become acidic. Record the number of drops of acid needed to generate the color change. Students should see that acid added to water or NaCl immediately changes the pH of these solutions rather significantly; however it takes quite a bit of acid to change the pH of the buffered solution.

### 4. Commercial Antacids

This procedure is designed to test the buffering effects of different commercial antacids. That is, it will test how well they can absorb excess $H^+$ ions, thus "neutralize stomach acid." The stomach produces HCl; therefore, it is the acid of choice for this experiment. Again, Brom Cresol Purple is used as a color indicator to show when the buffer is no longer neutralizing the acid.

The following products should be tested:

Rolaids®
Tums®
Maalox®
Alka-Seltzer®
Distilled water

A tube should be labeled for each of the products listed above. Each student or group can prepare each product, or the instructor may prepare them in advance. Crush the amount needed for one dose of each medication and dilute each with distilled water to 100 mL. Some of the products may require extensive mixing to get most or all of the powder into solution.

Pipet 5 mL of the dissolved products into the appropriate tubes. Add about 4 drops of Brom Cresol Purple to each tube and mix until homogeneous.

Add 0.1 M HCl dropwise to one tube at a time and mix after each drop. Continue this process until the solution turns yellow, indicating an acidic solution. Record the number of drops of acid needed to generate the color change.

You should find that Alka-Seltzer absorbs the most acid, and Tums places second. You might want to explain to the students, however, that Alka-Seltzer contains sodium bicarbonate, while Tums consists of calcium carbonate. Thus, although Alka-Seltzer is most effective, people on sodium-restricted diets should avoid this and turn to Tums instead.

## Reference


Martini, F. (1989). *Fundamentals of anatomy and physiology*. New Jersey: Prentice Hall.