

Experiment GB-1: Biological Buffers

Exercise 1: The Effect of Adding 0.1M HCl to Water

Aim: To determine the changes in pH that take place in deionized water treated with a weak acid.

Procedure

1. Using the equipment from the calibration exercise, place 50 mL of room temperature deionized (DI) water in a clean 100 ml beaker. Add a stir bar to the beaker and place the beaker on the magnetic stirrer. Turn on the stirrer and position the stir bar to one side of the beaker bottom.
2. Remove the ISE-100 pH electrode from the beaker of deionized water used at the end of calibration. Blot the drops of DI water from the electrode. Mount the electrode in a clamp on the ringstand and position it over the new beaker of deionized water. Carefully lower the tip of the electrode into the beaker.
3. Turn on the stirrer so that the stir bar rotates slowly and evenly.
4. Click Record on the LabScribe Main window to begin recording. When the recording on the channel reaches a stable baseline, type DI Water in the Mark box to the right of the Mark button. Press the Enter key on the keyboard to mark the recording.
5. After recording for at least fifteen seconds of stable baseline, type 0.1M HCl-10 Drops on the Mark box.
6. As you add ten drops of 0.1M HCl to the beaker of deionized water, press the Enter key on the keyboard to mark the recording. Continue recording.
7. Type Add10 Drops on the Mark box. When the pH of the water in the beaker has reached a new stable level, add ten more drops of 0.1M HCl to the DI water. As you add the drops, press the Enter key on the keyboard to mark the recording. Continue recording.
8. Repeat Step 7 until a total of 60 drops of 0.1M HCl have been added to the DI water in the beaker.
9. Select Save in the File menu.
10. Turn off the magnetic stirrer. Remove the pH electrode from the beaker. Hold the electrode over the beaker used for collecting waste liquid, and rinse it with deionized water from a wash bottle. Blot any extra drops of DI water from the electrode and place it in a clean, empty beaker. The electrode will be used in the next exercise.
11. Remove the stir bar from the beaker of acidified water and rinse it with deionized water from a wash bottle. Discard the acidified water in the proper manner, and clean and rinse the beaker.

Data Analysis

1. Scroll to the section of data file in which the pH changes of deionized water treated with 0.1M HCl were recorded ([Figure GB-1-L1](#)).



Figure GB-1-L1: Recording of the change in pH of the water when HCL is added.

2. Use the Display Time icons on the LabScribe toolbar to position the complete recording on the Main window. The required data can also be selected by:
 - Placing the cursors on either side of the section of data needed. Place one cursor on the stable pH level recorded from pure deionized water. Place the second cursor on the stable pH level recorded after a total of 60 drops of 0.1M HCl were added to the deionized water
 - Clicking the Zoom between Cursors button on the LabScribe toolbar to expand the segment of data to the width of the Main window.
3. Click on the Analysis window icon in the toolbar or select Analysis from the Windows menu to transfer the data displayed in the Main window to the Analysis window ([Figure GB-1-L2](#)).
4. Look at the Function Table that is above the uppermost channel displayed in the Analysis window. The mathematical functions that are listed should include Title, Value1, Value2, and V2-V1. The values for these parameters from each channel are seen in the table across the top margin of each channel.
5. **5** Once the cursors are placed in the correct positions for determining the pH, the values for pH can be recorded in the on-line notebook of LabScribe by typing the names and values directly into the Journal.



Figure GB-1-L2: pH recording in the Analysis window showing the pH values for the addition of 0.1M HCl to the deionized water.

6. The functions in the channel pull-down menus of the Analysis window can also be used to enter the names and values of the parameters from the recording to the Journal. To use these functions:
 - Place the cursors at the locations used to measure the pH from the pH channel.
 - Transfer the name of the mathematical function used to determine the pH to the Journal using the Add Title to Journal function in the pH channel pull-down menu.
 - Transfer the value for the pH to the Journal using the Add Ch. Data to Journal function in the pH channel pull-down menu.
7. Place one cursor on the stable pH level of the pure deionized water. Place the second cursor on the stable pH level that occurs after the initial ten drops of 0.1M HCl were added to the deionized water.
8. Measure the values for the following parameters from the pH channel for the region of data selected:
 - pH-DI Water, which is Value1 on the pH channel.
 - pH-DI Water & 10 Drops of 0.1M HCl, which is Value2 on the pH channel.
9. Record the values for these parameters in the Journal using one of the procedures described in Step 6, and in [Table GB-1-L1](#).
10. Move the second cursor from the pH level of DI water containing ten drops of 0.1M HCl to the stable pH level of DI water containing 20 drops of 0.1M HCl, and repeat Steps 8 and 9 to measure and record the pH of the water after a total of twenty drops of 0.1M HCl were added to the water.
11. Move the second cursor to each of the stable pH levels of DI water containing 30, 40, 50, and 60 drops of 0.1M HCl, and repeat Steps 8 and 9 to measure and record the pH of the water after each addition of 0.1M HCl.
12. While the second cursor is on the pH level of DI water containing sixty drops of 0.1M HCl,

measure the Overall Change in pH using the parameter, V2-V1, from the Function Table in the Analysis window.

13. Divide the Overall Change in pH by the initial pH of the DI water to determine the percent change in pH.
14. Click Save in the File menu.

Exercise 2: The Effect of Adding 0.1M HCl to a Physiological Buffer

Aim: To determine the changes in pH that take place in a physiological buffer treated with a weak acid.

Procedure

1. Repeat Exercise 1 using a physiological buffer in place of deionized water.
2. Mark the recording with appropriate labels to indicate the number of drops of 0.1M HCl added to the buffer.
3. Select Save in the File menu to add this data to the existing data file.

Data Analysis

1. Use the same techniques used in Exercise 1 to measure the pH levels of the buffer after different amounts of 0.1M HCl were added.
2. Use the same techniques explained in Exercise 1 to record the values of the pH levels in the Journal, and in [Table GB-1-L1](#).

Exercise 3: The Effect of Adding 0.1M HCl to Common Solutions

Aim: To determine the changes in pH that take place in different solutions treated with a weak acid. Solutions that are better buffers will have smaller changes in pH.

Procedure

1. Repeat Exercise 1 using a solution which is assigned to your lab group, in place of deionized water. The groups in your lab session will be assigned a solution to test as a buffer. These may include:
 - Apple juice, 2% milk, Sports drink, Clear soft drink, or any other solution your instructor deems appropriate.
2. Mark the recording with corresponding labels to indicate the type of solution used and the number of drops of 0.1M HCl added to the solution.
3. Select Save in the File menu to add this data to the existing data file.

Data Analysis

1. Use the same techniques used in Exercise 1 to measure the pH levels of the solutions after different amounts of 0.1M HCl were added.
2. Use the same techniques explained in Exercise 1 to record the values of the pH levels in the Journal, and in the data table.

Exercise 4: The Effect of Adding 0.1M NaOH to Water

Aim: To determine the changes in pH that take place in deionized water treated with a weak base.

Procedure

1. Repeat Exercise 1 using 0.1M NaOH in place of 0.1M HCl.
2. Mark the recording with appropriate labels to indicate the number of drops of 0.1M NaOH added to the water.
3. Select Save in the File menu to add this data to the existing data file.

Data Analysis

1. Use the same techniques used in Exercise 1 to measure the pH levels of the DI water after different amounts of 0.1M NaOH have been added.
2. Use the same techniques explained in Exercise 1 to record the values of the pH levels in the Journal, and in [Table GB-1-L2](#).

Exercise 5: The Effect of Adding 0.1M NaOH to a Physiological Buffer

Aim: To determine the changes in pH that take place in a physiological buffer treated with a weak base.

Procedure

1. Repeat Exercise 1 using a physiological buffer in place of deionized water.
2. Mark the recording with appropriate labels to indicate the number of drops of 0.1M NaOH added to the buffer.
3. Select Save in the File menu to add this data to the existing data file.

Data Analysis

1. Use the same techniques used in Exercise 1 to measure the pH levels of the buffer after different amounts of 0.1M NaOH were added.
2. Use the same techniques explained in Exercise 1 to record the values of the pH levels in the Journal, and in [Table GB-1-L2](#).

Exercise 6: The Effect of Adding 0.1M NaOH to Common Solutions

Aim: To determine the changes in pH that take place in different solutions treated with a weak base. Solutions that are better buffers will have smaller changes in pH.

Note: Some solutions will be better at buffering an acid than a base and visa versa. Note this while observing your results and those of the other lab groups.

Procedure

1. Repeat Exercise 1 using the same solution that was assigned to your lab group when testing an acid, in place of deionized water.
2. Mark the recording with appropriate labels to indicate the type of solution used and the number of drops of 0.1M NaOH added to the solution.
3. Select Save in the File menu to add this data to the existing data file.

Data Analysis

1. Use the same techniques used in Exercise 1 to measure the pH levels of the solutions after different amounts of 0.1M NaOH were added.
2. Use the same techniques explained in Exercise 1 to record the values of the pH levels in the Journal, and in the data table.

Table GB-1-L12: Changes in pH in Deionized Water, Apple Juice, 2% Milk, and Other Solutions Treated with 0.1M HCl.

0.1M HCl	Deionized Water		Apple Juice		2% Milk		Sports Drink		Colorless Soft Drink		Buffered Phys. Saline	
	pH	Overall ΔpH	pH	Overall ΔpH	pH	Overall ΔpH	pH	Overall ΔpH	pH	Overall ΔpH	pH	Overall ΔpH
Solution Only												
Solution & 10 Drops												
Solution & 20 Drops												
Solution & 30 Drops												
Solution & 40 Drops												
Solution & 50 Drops												
Solution & 60 Drops												
Percent Change												

Table GB-1-L2: Changes in pH in Deionized Water, Apple Juice, 2% Milk, and Other Solutions Treated with 0.1M NaOH.

0.1M NaOH	Deionized Water		Apple Juice		2% Milk		Sports Drink		Colorless Soft Drink		Buffered Saline	
	pH	Overall ΔpH	pH	Overall ΔpH	pH	Overall ΔpH	pH	Overall ΔpH	pH	Overall ΔpH	pH	Overall ΔpH
Solution Only												
Solution & 10 Drops												
Solution & 20 Drops												
Solution & 30 Drops												
Solution & 40 Drops												
Solution & 50 Drops												
Solution & 60 Drops												
Percent Change												

Questions

- Besides the buffered physiological saline, which solution demonstrated the lowest percent change in its pH when treated with a weak acid?
- Besides the buffered physiological saline, which solution demonstrated the lowest percent change in its pH when treated with weak base? Is it the same solution as the one that responds with the lowest percent change in pH when treated with a weak acid?

3. Which solution demonstrated the highest percent change in its pH when treated with a weak acid?
4. Which solution demonstrated the highest percent change in its pH when treated with weak base? Is it the same solution as the one that responds with the highest percent change in pH when treated with a weak acid?
5. Besides the buffered saline, which solutions would be good buffers? Are all these solutions biological?
6. Why would each solution that you selected be a good buffer?