Experiment GB-2: Membrane Permeability

Equipment Required

PC or Mac Computer

IXTA, USB cable, IXTA power supply

ISE-100 combination pH electrode

Magnetic stirrer and stir bar

Ringstand and clamps

100 ml beakers (2)

250 ml beakers (6)

1000 ml beaker

pH 4 and pH 7 buffer solutions

1.0M HCl solution

1.0M NaOH solution

1.0M Na Acetate solution

2 cm wide dialysis tubing

String

Medium Binder Clips (3)

Deionized water

pH Electrode Setup

1. Locate the ISE-100 pH electrode andplug the DIN8 connector into the Channel A5.

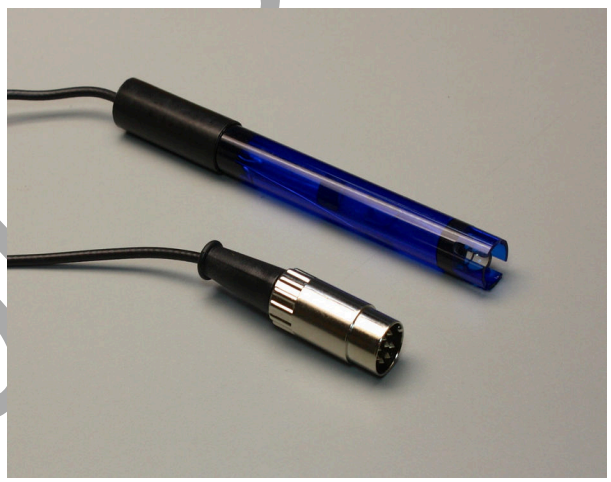

Figure GB-2-S1: The ISE-100 pH electrode.

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Calibration of the pH Electrode

1. If the ISE-100 pH electrode is still stored in its bottle of buffer, remove the electrode from the bottle. Rinse the electrode with deionized water while holding the electrode over a 1000 ml beaker used for the collection of waste liquids.
2. Place the tip of the ISE-100 pH electrode in a 100 ml beaker containing enough room temperature deionized water to submerge the tip. Keep the electrode in deionized water for at least ten minutes.

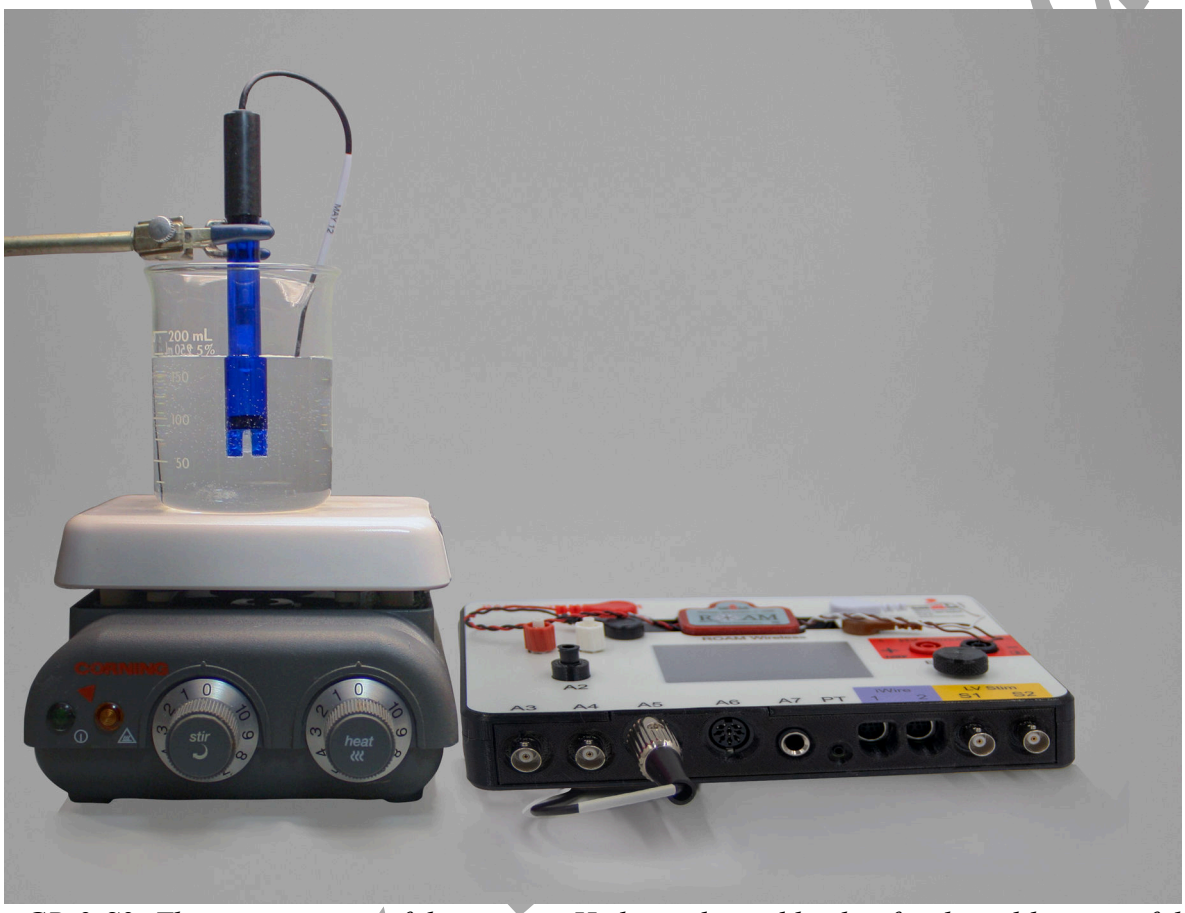

Figure GB-2-S2: The arrangement of the stirrer, pH electrode, and beaker for the calibration of the pH probe.

3. Prepare two 100 ml beakers, each filled with 50 ml of the pH buffers used for calibrating the pH electrode. The buffers should be at room temperature. One beaker is filled with pH 7 buffer; and the other is filled with pH 4 buffer. Each beaker should be filled with enough buffer to cover the tip of the ISE-100 pH electrode, and also allow the stir bar in the beaker to spin without touching the electrode.
4. Place the beaker containing the pH 7 buffer on the magnetic stirrer. Carefully place a stir bar in the beaker. Remove the electrode from the deionized water and blot any extra drops of water. Position the tip of the electrode in the beaker of pH 7 buffer so that the tip is away from the stir bar. Adjust the speed of the stirrer so the stir bar is rotating evenly at a slow speed.

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5. Click Record. After a few seconds, the trace will reach a stable baseline toward the top of the recording channel. Type the words **Calibration - pH 7** in the Mark box. Click the mark button to mark the stable baseline of the recording. This will mark the output of the ISE-100 pH electrode in pH 7 buffer. Continue recording while changing the beakers of buffers.
6. Turn off the stirrer and remove the ISE-100 pH electrode from the beaker of pH 7 buffer. Hold the electrode over the beaker used for collecting waste liquid and rinse it with deionized water. Blot any extra drops of water.
7. Place the beaker of pH 4 buffer on the stirrer. Carefully place a stir bar in the beaker. Position the tip of the electrode in the beaker of pH 4 buffer so that it is away from the stir bar. Adjust the speed of the stirrer so the stir bar is rotating evenly at a slow speed.
8. As you continue to record, the trace will reach a stable baseline toward the bottom of the recording channel. Type the words **Calibration - pH 4** in the Mark box. Click the mark button to mark the stable baseline of the recording.
9. Click Stop to halt the recording.
10. Select Save As in the File menu, type a name for the file. Click on the Save button to save the data file.
11. Turn off the stirrer. Remove the electrode from the beaker of pH 4 buffer. 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 water and place the electrode in a beaker of deionized water.

Units Conversion

1. Scroll to the beginning of the calibration data for the ISE-100 pH electrode.
2. Use the Display Time icons to adjust the Display Time of the Main window to show the data collected at pH 7 and pH 4on the Main window at the same time.
3. Click the 2-Cursor icon so that two cursors appear on the Main window. Place one cursor on the flat section of data collected when the ISE-100 pH electrode was in the pH 7 buffer and the second cursor on the flat section of data collected when the electrode was in the pH 4 buffer.
4. To convert the voltages at the positions of the cursors to pH values, use the Simple Units Conversion dialogue window. Click on V2-V1 to the right on the pH channel and select Simple.
5. On the units conversion window, make sure 2 point calibration is selected in the pull-down menu in the upper-left corner of the window. Put a check mark in the box next to Apply units to all blocks. Notice that the voltages from the positions of the cursors are automatically entered into the value equations. Enter the two buffers used in the calibration recording in the corresponding boxes on the right side of the conversion equations. Click OK.

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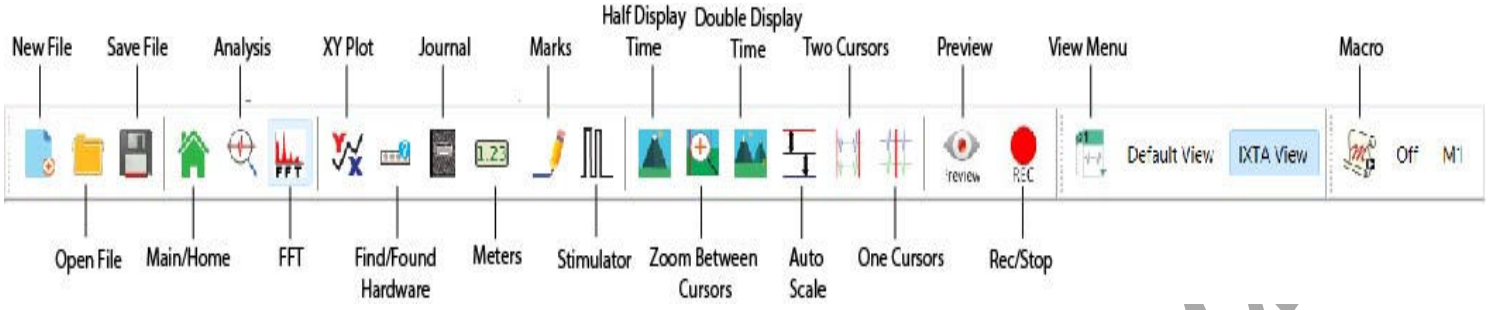

Figure GB-2-S3: The LabScribe toolbar.

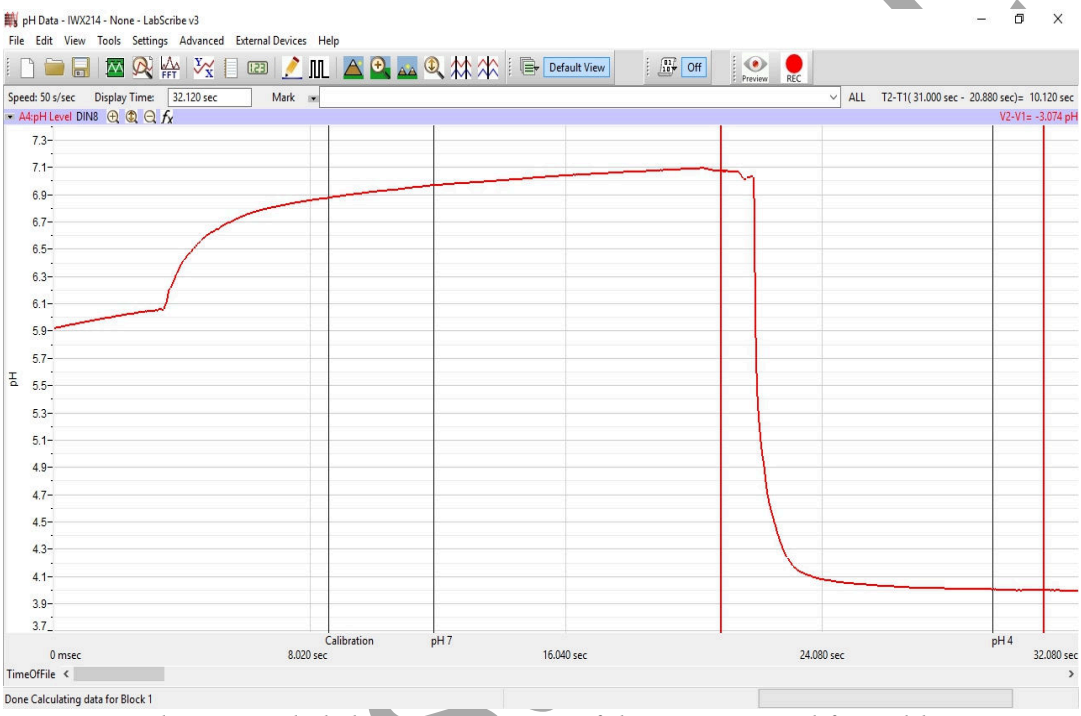

Figure GB-2-S4: pH data recorded showing positions of the cursors used for calibration.

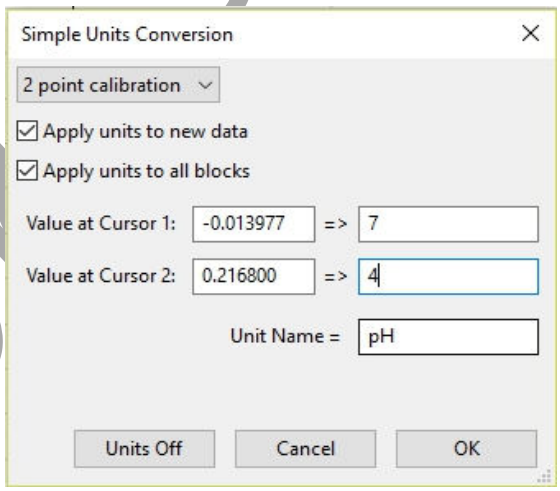

Figure GB-2-S5: The Simple Units Conversion dialogue window.

Work

Preparation of the Dialysis Tubes

1. Obtain three pieces of two centimeter-wide dialysis tubing that are fifteen centimeters long. Soak the tubing in deionized water to soften them.
2. Knot one end of each dialysis tubing to form a sac. The other end will be tied with string after filling.
3. Fill one of the sacs about two-thirds full with 1.0M HCl. Carefully twist the bag closed, trying to remove all air bubbles. Tie the end tightly with string to prevent any leakage.
4. Rinse the outside of the dialysis sac containing the 1.0M HCl with tap water to remove any acid that may have gotten on the outside.
5. Stand the completed dialysis sac with 1.0M HCl in a clean, dry 250 ml beaker. Label the beaker.
6. Repeat Steps 3, 4, and 5 to make a sac filled with 1.0M NaOH and a sac filled with 1.0M Na Acetate. Put each dialysis sac in its own clean, dry 250 ml beaker.

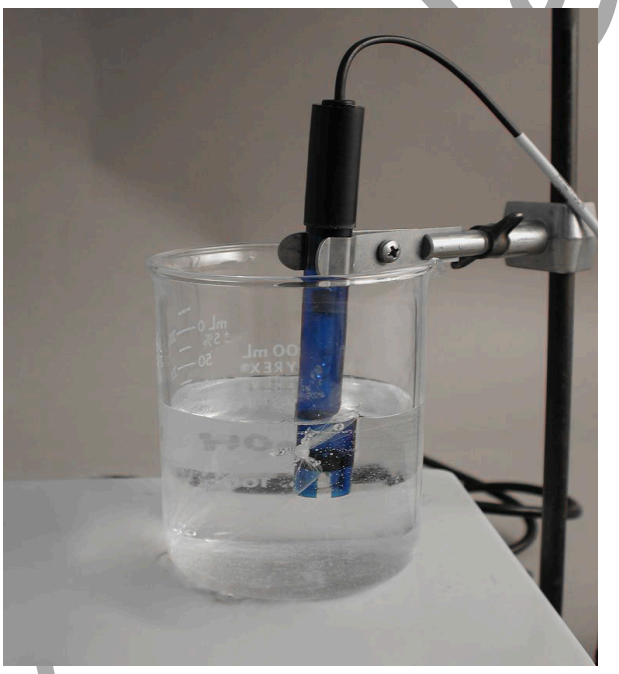

Figure GB-2-S6: View of the dialysis tubing in the beaker with the ISE-100 pH electrode.

IworX Science Lab

Experiment GB-2: Membrane Permeability

Exercise 1: Movement of Small Positive Ions Across a Membrane

Aim: To determine if small, positively charged, hydrogen ions can move across a membrane from a region of higher concentration (1.0M HCl) to a region of lower concentration (deionized water).

Approximate Time: 15 minutes

Procedure

1. Using the equipment from the calibration exercise, place 100 mL of room temperature deionized (DI) water in a clean 250 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. Click the mark button to mark the recording.
5. After recording at least fifteen seconds of stable baseline, type the words **Dialysis Tube w/ 1.0M HCl** in the Mark box.
6. Lower the dialysis tube into the deionized water and click the mark button to mark the recording. Clamp the dialysis tube to the edge of the beaker so that about half the solution in the tube is below the surface of the deionized water. Make sure the bottom of the dialysis tube is not touching the stir bar.
7. Record the pH of the deionized water for five minutes.
8. Mark the recording at the 1 minute interval, type **1 minute** in the Mark box and click the mark button to mark the recording.
9. Repeat Step 8 for the 2, 3, 4, and 5 minute intervals.
10. At the end of five minutes, click Stop to halt the recording.
11. Select Save in the File menu.
12. Turn off the magnetic stirrer. Remove the dialysis tube with the 1.0M HCl from the beaker and discard the dialysis sac as directed.
13. 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 drops of water from the electrode and place it in a beaker of deionized water.
14. Remove the stir bar from the beaker of deionized water and rinse it with deionized water from a wash bottle. Discard the deionized water.

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Data Analysis

1. Scroll to the section of data recorded in which the pH changes of deionized water treated with the 1.0M HCl dialysis tubing were recorded as shown below.

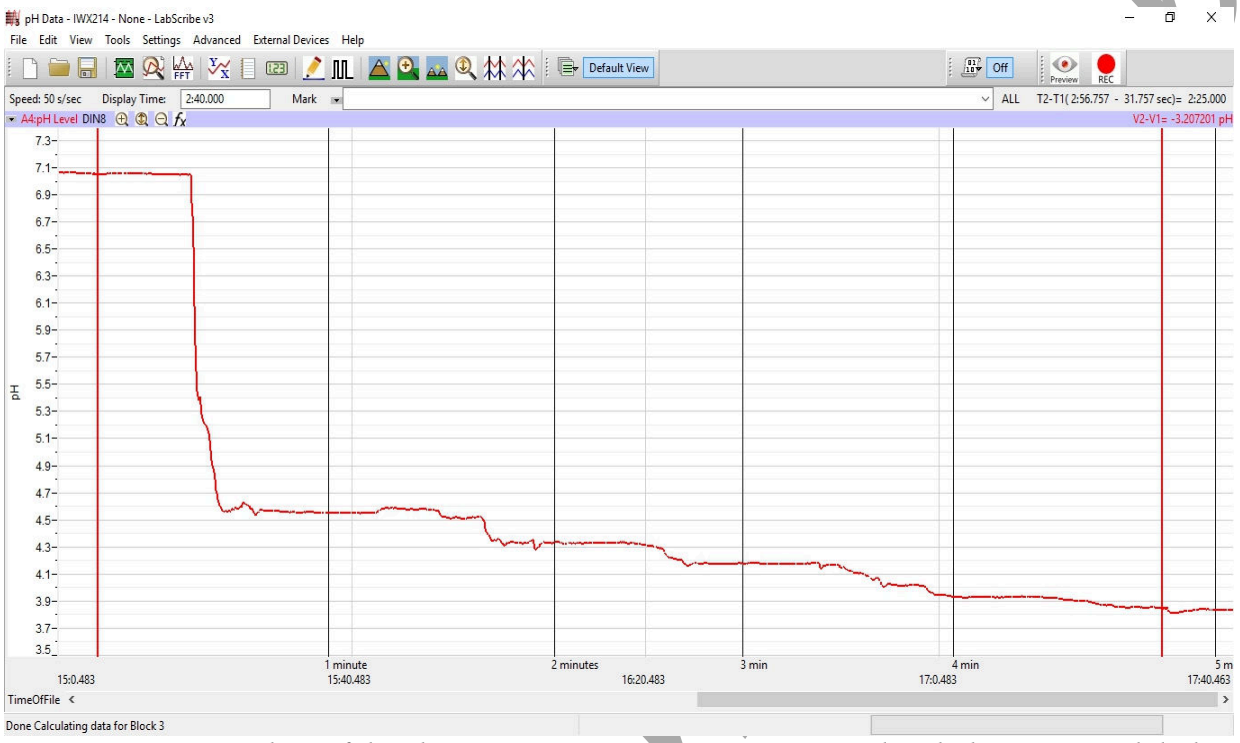

Figure GB-2-L1: Recording of the changes in pH over a 5 minute period with the 1.0 M HCl dialysis tubing in deionized water.

2. Use the Display Time icons on the LabScribe toolbar to position the complete recording on 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.
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. 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.

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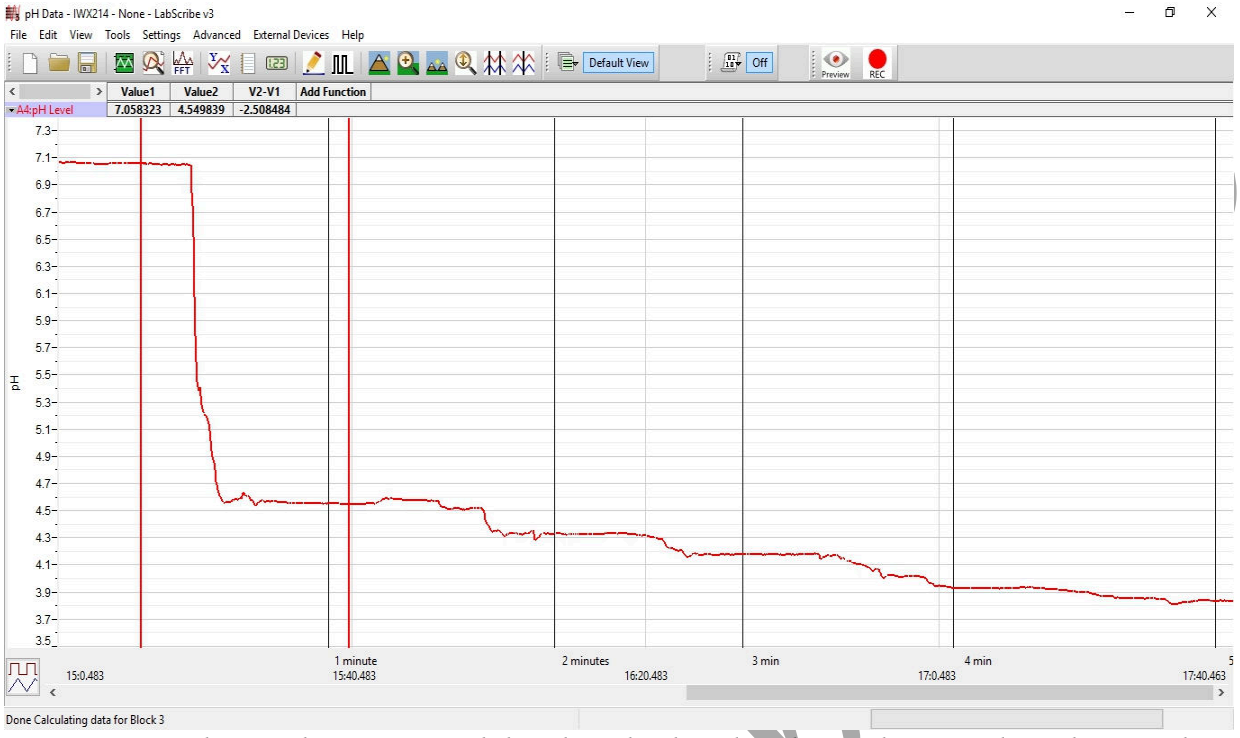

Figure GB-2-L2: The membrane permeability data displayed in the Analysis window, showing the values at each cursor and the pH change over time with the addition on a dialysis tube of 1.0 M HCl.

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 a cursor on the stable baseline recorded just before the dialysis tubing containing 1.0M HCl was lowered into the beaker of deionized water. Place the second cursor at the point in the recording that is five minutes after the dialysis tubing was placed in the 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.
    - 5 minutes, 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 1.
10. Measure the Overall Change in pH using the parameter, V2-V1, from the Function Table in the Analysis window.

Work Sample

11. Divide the Overall Change in pH by the initial pH of the DI water, and multiply by 100 to determine the percent change in pH.
12. Click Save in the File menu.

Exercise 2: Movement of Small Negative Ions Across a Membrane

Aim: To determine if small, negatively charged, hydroxide ions can move across a membrane from a region of higher concentration (1.0M NaOH) to a region of lower concentration (deionized water).

Approximate Time: 15 minutes

Procedure

1. Repeat Exercise 1 using the 1.0M NaOH dialysis sac in place of the 1.0M HCl.
2. Mark the recording with appropriate labels to indicate the length of time the 1.0M NaOH dialysis sac was in the beaker of deionized water.
3. Select Save in the File menu to add this data to the existing data file. Remember to remove the pH electrode and rinse well with deionized water.

Data Analysis

1. Use the same techniques used in Exercise 1 to measure the pH levels of the DI water after the 1.0M NaOH dialysis sac was immersed in the deionized water.
2. Use the same techniques explained in Exercise 1 to record the values of the pH levels in the Journal, and in Table 1.
3. Click Save in the File menu.

Exercise 3: Movement of Large Ions Across a Membrane

Aim: To determine if large, negatively charged, acetate ions can move across a membrane from a region of higher concentration (1.0M Na Acetate) to a region of lower concentration.

Approximate Time: 15 minutes

Procedure

1. Repeat Exercise 1 using the 1.0M Na Acetate dialysis sac in place of the 1.0M HCl.
2. Mark the recording with appropriate labels to indicate the length of time the 1.0M Na Acetate dialysis sac was in the beaker of deionized water.
3. Select Save in the File menu to add this data to the existing data file.

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Data Analysis

1. Use the same techniques used in Exercise 1 to measure the pH levels of the DI water after the 1.0M Na Acetate dialysis sac was immersed in the deionized water.
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.
3. Click Save in the File menu.

Table GB-2-L1: Changes in pH during Movement of Ions.

Solution in Dialysis Tubing	pH Level		
	T=0 Mins	T=5 Mins	Change (Δ)
1.0M HCl			
1.0M NaOH			
1.0M Na Acetate			

Questions

1. During the five minutes that the dialysis sac containing 1.0M HCl was placed in the beaker of deionized water, what happened to the pH of the water? What caused the result that you recorded?
2. During the five minutes that the dialysis sac containing 1.0M NaOH was placed in the beaker of deionized water, what happened to the pH of the water? What caused this result?
3. During the five minutes that the dialysis sac containing 1.0M Na Acetate was placed in the beaker of deionized water, what happened to the pH of the water?
4. Explain how the movement of large particles causes a change in pH.
5. If any of the ions diffused across the membrane, which one diffused more quickly?
6. What factors could increase the rate of diffusion of an ion across a membrane?

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