



**University of Al-Ameed
College of Pharmacy**

2nd stage , 1st Semester



Practical Physical Pharmacy

Buffer Solution/ Lab 6

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Buffer solution:

- Is a solution that **resist changes in pH** when small quantities of acid or alkali are added.

Uses of Buffer:

1. Uses of Buffers in Biological Systems:

- **Saliva:** The pH of saliva typically ranges from 6.2 to 7.6, which is essential for maintaining oral health. Buffers in saliva, primarily bicarbonate and phosphate ions, help neutralize acids produced by bacteria during the digestion of carbohydrates. This buffering action protects tooth enamel and maintains an environment conducive to oral flora.

- **Blood:** Blood maintains a pH range of 7.35 to 7.45, crucial for proper physiological functions. The bicarbonate buffer system is the primary mechanism that regulates blood pH. When excess carbon dioxide (CO_2) is produced during metabolism, it forms carbonic acid (H_2CO_3), which dissociates into bicarbonate (HCO_3^-) and hydrogen ions (H^+). This buffer system helps prevent dangerous fluctuations in pH that could lead to conditions like acidosis or alkalosis.

2. Uses of Buffers in Cosmetics and Personal Hygiene Products:

- **pH Stability:** Buffers are crucial in formulating cosmetic and personal care products, such as shampoos, lotions, and creams, to maintain a neutral or slightly alkaline pH. This helps prevent skin irritations that can occur when products are too acidic or too alkaline.
- **Topical Products:** Ointments and creams often contain buffers to ensure product stability over time. Common buffering agents include citric acid and its salts, as well as phosphoric acid, which help maintain the desired pH for efficacy and skin compatibility.

There are two types of buffer solution:

- 1. Acidic Buffer**
- 2. Basic Buffer**

Acidic Buffer :

- Definition:** Acidic buffers consist of a weak acid and its conjugate base (salt).
- Example:** Acetate buffer (CH_3COOH and CH_3COONa). **Acetic acid** (CH_3COOH) as a weak acid and **sodium acetate (CH_3COONa)** as its salt.

Note: The acetate buffer maintains a stable pH around 4.75, which is ideal for many biochemical processes and laboratory applications requiring an acidic environment.

Basic Buffer:

- **Definition:** Basic buffers consist of a weak base and its conjugate acid (salt).
- **Example:** Ammonium buffer or ammonia buffer ($\text{NH}_4\text{OH} + \text{NH}_4\text{Cl}$).
Ammonium hydroxide (NH_4OH) as the weak base and ammonium chloride (NH_4Cl) as its salt.

Note: The ammonia buffer is well-known for maintaining a stable pH around 9.25, which is often required in biochemical and analytical applications.

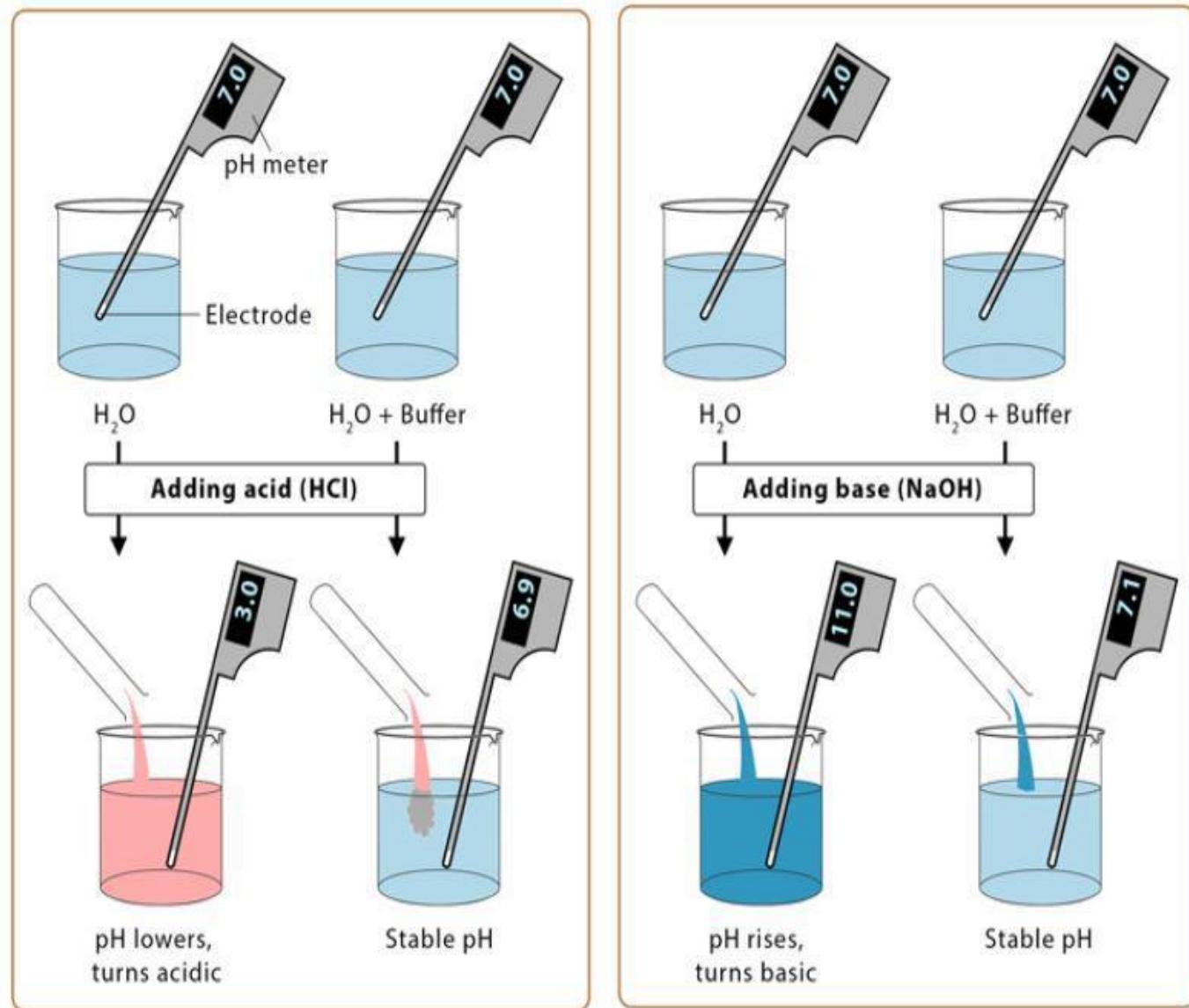
Phosphate Buffer (Sorensen's Buffer Solution):

- The phosphate buffer consists of **Sodium dihydrogen phosphate** (NaH_2PO_4) which acts as the acid component, and **Disodium hydrogen phosphate** (Na_2HPO_4) which acts as the conjugate base (salt) component.
- **pH Range:** The phosphate buffer is effective across a broad pH range, generally around pH 6.8-7.4. By adjusting the ratio of NaH_2PO_4 to Na_2HPO_4 , the pH can be precisely controlled.
- **Applications:** This buffer is extensively used in biological and medical fields due to its compatibility with biological systems.

Differentiation between buffer system & non-buffer system:

For example:

- If 1 ml of **0.1N HCl** solution is added to 100ml of **pure water**, the pH is reduced from 7 to 3.
- When **strong acid** is added to 1M solution containing equal quantities of **acetic acid & sodium acetate** the pH change only by 0.09 units because the base CH_3COO^- ties up the H^+ ion according to the following equation:
$$\text{CH}_3\text{COO}^- + \text{H}^+ \rightarrow \text{CH}_3\text{COOH} + \text{H}_2\text{O}$$



Mechanism of buffer action:

Consider a buffer solution that consists of a weak acid and its salt such as the acetate buffer:



- **Addition of Acid (H^+):**

When a strong acid (e.g. HCl) is added, it will be consumed by the conjugate base, forming the weak acid: $\text{CH}_3\text{COONa} + \text{HCl} \rightarrow \text{CH}_3\text{COOH} + \text{NaCl}$. This reaction consumes some of the added H^+ ions, minimizing the change in pH.

- **Addition of Base (OH^-):**

When a strong base (e.g. NaOH) is added, it will be consumed by the weak acid, forming the conjugate base: $\text{CH}_3\text{COOH} + \text{NaOH} \rightarrow \text{H}_2\text{O} + \text{CH}_3\text{COONa}$. This reaction reduces the concentration of OH^- ions and again helps to stabilize the pH.

pH Measurement



pH value can be measured using:

1. **Mathematical method:** by using the Henderson-Hasselbalch equation

- Henderson-Hasselbalch equation for a weak acid and its salt:

$$\text{pH} = \text{pK}_a + \log \frac{[\text{Salt}]}{[\text{Acid}]}$$

- Henderson-Hasselbalch equation for a weak base and its salt:

$$\text{pH} = \text{pK}_w - \text{pK}_a + \log \frac{[\text{Base}]}{[\text{Salt}]}$$

2. **Electrometric method:** by using pH meter.

3. Colorimetric method:

It is less accurate and less convenient but less expensive than the electronic method.

Colorimetric method involves the use of:

- a. **Paper indicator:** a strip of paper that undergo color changes depending on the pH of the solution where the strip is immersed.
- b. **liquid indicator:** they are weak acid or weak base that show color changes as their degree of dissociation varies with pH. The color of the indicator resembles a certain pH value.
 - **An examples of liquid indicators:** methyl yellow, methyl red, bromothymol blue, thymol blue, and phenolphthalein.

- Examples of calculating the pH of a buffer solution using Henderson-Hasselbalch equation:

Example 1. Calculate the pH of a buffer solution that is **0.050 M** in benzoic acid and **0.150 M** in sodium benzoate, **pka = 4.2**.

$$\text{pH} = \text{pka} + \log \frac{[\text{salt}]}{[\text{acid}]}$$

$$\text{pH} = 4.2 + \log \frac{0.150}{0.050}$$

$$\text{pH} = 4.2 + \log 3$$

$$\text{pH} = 4.2 + 0.48 = 4.68$$

Example 2. A buffer is prepared containing **1.00 M** acetic acid and **1.00 M** sodium acetate. What is its pH? the **pKa = 4.76**.

$$\text{pH} = \text{pKa} + \log \frac{[\text{salt}]}{[\text{acid}]}$$

$$\text{pH} = 4.76 + \log 1/1$$

$$\text{pH} = 4.76 + \log 1$$

$$\text{pH} = 4.76 + 0 = 4.76$$

Example 3: A buffer is prepared containing **0.800 molar** acetic acid and **1.00 molar** sodium acetate. What is its pH? **pKa = 4.76.**

$$\text{pH} = \text{pka} + \log \frac{[\text{salt}]}{[\text{acid}]}$$

$$\text{pH} = 4.76 + \log \frac{1}{0.8}$$

$$\text{pH} = 4.76 + \log 1.25$$

$$\text{pH} = \mathbf{4.76 + 0.09 = 4.85}$$

Example 4: A buffer is prepared containing **1.00 molar** acetic acid and **0.800 molar** sodium acetate. What is its pH? the pKa of acetic acid = 4.76

$$\text{pH} = \text{pka} + \log \frac{[\text{salt}]}{[\text{acid}]}$$

$$\text{pH} = 4.76 + \log \frac{0.8}{1}$$

$$\text{pH} = 4.76 + \log 0.8$$

$$\text{pH} = \mathbf{4.76 - 0.09 = 4.67}$$

Buffer capacity:

Refers to the ability of a buffer solution to resist changes in pH when strong acids or bases are added. The buffer capacity is numerically expressed as the minimum concentration of strong acid or base required to cause a one-unit change in pH.

- **To calculate buffer capacity, we use the following formula:**

$$\beta = \Delta B / \Delta pH$$

Where:

- β is a buffer capacity (which is unitless) .
- ΔB is the amount of strong acid or strong base added (the concentration in a normality 'N').
- ΔpH is the pH difference between the initial buffer's pH and the pH after the addition of acid or base to the buffer.

Examples of Buffer Capacity

- **Example 1:** 0.5 ml of 0.1 M NaOH is added to 40 ml of buffer solution having pH 3.75 and the pH will be 3.78 after the addition, **calculate the buffer capacity?**

- **Solution:** $\beta = \Delta B / \Delta pH$

First, we find ΔB using the following formula: $C_1V_1 = C_2V_2$

Since, 0.1 M NaOH = 0.1 N NaOH, because the Eq. wt = M.wt

$$0.1 N \times 0.5 \text{ ml} = C_2 \times (40 + 0.5) \text{ ml}$$

$\Delta B = C_2 = 0.0012 \text{ N}$ is the normality of NaOH in the final solution.

$$\Delta pH = 3.78 - 3.75 = 0.03$$

$$\beta = 0.0012 / 0.03 = 0.04$$

Thus, the buffer capacity of solution is **0.04**

- **Example 2:** We are given 60 mL of a sodium phosphate buffer with a pH of 7.39. We then add to it 150 mL of 0.2 M HCl. This addition gives the buffer solution a new pH of 7.03. What is the capacity of our sodium phosphate buffer?
- **Solution 1:** $\beta = \Delta B / \Delta \text{pH}$

$0.2 \text{ M HCl} = 0.2 \text{ N HCl}$, because the Eq. wt = M.wt

$$C_1 V_1 = C_2 V_2$$

$$0.2 \text{ N} \times 150 \text{ ml} = C_2 \times (60 + 150) \text{ ml}$$

$\Delta B = C_2 = 0.04 \text{ N}$ is the normality of HCl in the final solution

$$\Delta \text{pH} = 7.03 - 7.39 = 0.36$$

$$\beta = 0.04 / 0.36 = 0.11$$

Thus, the buffer capacity of solution is **0.11**

Solution 2:

- First, we find ΔB by dividing the number of moles of HCl we added to the buffer by the initial volume of the buffer. Don't forget your units, should be in liters!

Number of moles of HCl = $0.2 \text{ M} \times 0.150 \text{ L} = 0.03 \text{ mole}$

$$\Delta B = 0.03 \text{ mol} / 0.750 \text{ L} = \mathbf{0.04 \text{ M}}$$

$$\Delta \text{pH} = 7.03 - 7.39 = \mathbf{0.36}$$

$$\beta = 0.04 / 0.36 = \mathbf{0.11}$$

Thus, the buffer capacity of our sodium phosphate solution is **0.11**

- **Note 1:**

1. If a buffer has a **higher concentration of its components** (e.g. the weak acid and its conjugate base or a weak base and its conjugate acid), **its capacity will be higher**.
 - **For example:** a buffer with **[weak acid] = 0.50 M** and **[conjugate base] = 0.50 M** would have a higher capacity than a buffer with **[weak acid] = 0.050 M** and **[conjugate base] = 0.050 M**.
2. If a buffer has a **greater concentration of the weak acid** than that of the conjugate base, then it will have **a higher capacity for added base**. **For example**, in an acetate buffer with **0.1 M CH₃COOH** (weak acid) and **0.01 M CH₃COO⁻** (conjugate base), the higher weak acid concentration increases the buffer's capacity to resist changes in pH when a base is added.
3. Likewise, if a buffer has a **greater concentration of the conjugate base** than that of the weak acid, then it will have **a higher tolerance for added acid**.

Note 2:

1. Addition of **strong acid** to **acidic buffer solution** lead to increase the acid concentration and decrease the salt concentration.
$$\text{pH} = \text{pka} + \log \frac{[\text{salt} - \text{strong acid}]}{[\text{acid} + \text{strong acid}]}$$
2. Addition of **strong base** to **acidic buffer solution** lead to decrease the acid concentration and increase the salt concentration.
$$\text{pH} = \text{pka} + \log \frac{[\text{salt} + \text{strong base}]}{[\text{acid} - \text{strong base}]}$$
3. Addition of **strong base** to **basic buffer solution** lead to increase the base concentration and decrease the salt concentration.
4. Addition of **strong acid** to **basic buffer solution** lead to decrease the base concentration and increase the salt concentration.

1. Addition of **strong acid** to **acidic buffer solution** lead to increase the acid concentration and decrease the salt concentration. $pH = pKa + \log \frac{[\text{salt} - \text{strong acid}]}{[\text{acid} + \text{strong acid}]}$

Example: An acidic buffer is prepared containing **0.1 M** acetic acid and **0.1 M** sodium acetate. **0.01 M** of HCl has been added. Calculate the pH before and after HCl added? **pKa = 4.76.**

- **The pH before HCl added:**

$$pH = pKa + \log \frac{[\text{salt}]}{[\text{acid}]}$$

$$pH = 4.76 + \log \frac{0.1}{0.1}$$

$$pH = 4.76 + \log 1$$

$$pH = 4.76 + 0 = \mathbf{4.76}$$

- **The pH after HCl added:**

$$pH = pKa + \log \frac{[\text{salt} - \text{strong acid}]}{[\text{acid} + \text{strong acid}]}$$

$$pH = pKa + \log \frac{[0.1 - 0.01]}{[0.1 + 0.01]}$$

$$pH = 4.76 + \log \frac{0.09}{0.11}$$

$$pH = 4.76 + \log 0.82$$

$$pH = 4.76 - 0.09 = \mathbf{4.67}$$

2. Addition of **strong base** to **acidic buffer solution** lead to decrease the acid concentration and increase the salt concentration. $pH = pKa + \log \frac{[\text{salt} + \text{strong base}]}{[\text{acid} - \text{strong base}]}$

Example: An acidic buffer is prepared containing **0.10 M** acetic acid and **0.10 M** sodium acetate. **0.01 M** of NaOH has been added. Calculate the pH before and after NaOH added? **pKa = 4.76.**

- **The pH before NaOH added:**

$$pH = pKa + \log \frac{[\text{salt}]}{[\text{acid}]}$$

$$pH = 4.76 + \log \frac{0.1}{0.1}$$

$$pH = 4.76 + \log 1$$

$$pH = 4.76 + 0 = \mathbf{4.76}$$

- **The pH after NaOH added:**

$$pH = pKa + \log \frac{[\text{salt} + \text{strong base}]}{[\text{acid} - \text{strong base}]}$$

$$pH = pKa + \log \frac{[0.1+0.01]}{[0.1-0.01]}$$

$$pH = 4.76 + \log \frac{0.11}{0.09}$$

$$pH = 4.76 + \log 1.22$$

$$pH = 4.76 + 0.09 = \mathbf{4.85}$$

Example: An acidic buffer is prepared containing **0.1 M** acetic acid and **0.1 M** sodium acetate. **0.01 M** of NaOH has been added. Calculate the pH after NaOH added? **pKa = 4.76.**

$$\text{pH} = \text{pka} + \log \frac{[\text{salt} + \text{strong base}]}{[\text{acid} - \text{strong base}]}$$

$$\text{pH} = \text{pka} + \log \frac{[0.1 + 0.01]}{[0.1 - 0.01]}$$

$$\text{pH} = 4.76 + \log \frac{0.11}{0.09}$$

$$\text{pH} = 4.76 + \log 1.22$$

$$\text{pH} = 4.76 + 0.09 = \mathbf{4.85}$$

Example: An acidic buffer is prepared containing **0.05 M** acetic acid and **0.05 M** sodium acetate. **0.01 M** of NaOH has been added. Calculate the pH after NaOH added?

$$\mathbf{pKa = 4.76.}$$

$$\text{pH} = \text{pka} + \log \frac{[\text{salt} + \text{strong base}]}{[\text{acid} - \text{strong base}]}$$

$$\text{pH} = \text{pka} + \log \frac{[0.05 + 0.01]}{[0.05 - 0.01]}$$

$$\text{pH} = 4.76 + \log \frac{0.06}{0.04}$$

$$\text{pH} = 4.76 + \log 1.5$$

$$\text{pH} = 4.76 + 0.17 = \mathbf{4.93}$$



Experiment

- **Title:**
 - Preparation and Testing of a Acetate Buffer
- **Aim:**
 - To prepare acetate buffer solution that maintains a pH of approximately 4.75, suitable for biological and chemical applications.
 - To measure the pH of the prepared acetate buffer solution using different method
 - To measure and analyze the buffer capacity of a buffer solution against added acid or base.
- **Materials and equipment:**
 1. 0.1 M acetic acid, 0.1 M sodium acetate, and distilled water.
 2. Conical flasks, volumetric flasks, beaker , pipette, pH meter.

Procedure:

1. Preparation of acetate buffer solution

- Preparation of 1 L of 0.1 M Acetic acid

- a) To prepare a 0.1 M acetic acid solution, use the formula:

$$\text{Mass} = M \times M_{\text{wt}} \times V = 0.1 \times 60.06 \times 1 \text{ (liter)} = 6.006 \text{ g.}$$

- b) Use an analytical balance to accurately weigh out 6.006g of acetic acid.
 - c) Transfer the 6.006g of acetic acid into a 1-liter volumetric flask and add distilled water.
 - d) Stir until the solid dissolves completely.
 - e) Label the solution as "0.1 M acetic acid" and store it properly.

- **Preparation of 1 L of 0.1 M Sodium acetate**

- a) To prepare a 0.1 M sodium acetate solution, use the formula:

$$\text{Mass} = M \times M_{\text{wt}} \times V = 0.1 \times 136.06 \times 1(\text{liter}) = 13.61\text{g.}$$

- b) Use an analytical balance to accurately weigh out 13.61g of acetic acid.
 - c) Transfer the 13.61 g of sodium acetate into a 1-liter volumetric flask and add distilled water.
 - d) Stir until the solid dissolves completely.
 - e) Label the solution as "0.1 M sodium acetate" and store it properly.

- **Buffer Preparation:**

1. Prepare the following acetate buffer solutions by mixing specific volumes of 0.1 M acetic acid and 0.1 M sodium acetate, then diluting to a final volume of 100 mL with distilled water:
 - 25 ml of 0.1 M solution of acetic acid + 25 ml of 0.1 M solution of sodium acetate
 - 45 ml of 0.1 M solution of acetic acid + 5 ml of 0.1 M solution of sodium acetate
 - 5 ml of 0.2 M solution of acetic acid + 45 ml of 0.2 M solution of sodium acetate
 - 20 ml of 0.2 M solution of acetic acid + 30 ml of 0.2 M solution of sodium acetate
 - 30 ml of 0.2 M solution of acetic acid + 20 ml of 0.2 M solution of sodium acetate
2. Stir thoroughly to ensure complete mixing of the two solutions.

2. Measurement of pH:

The pH of each buffer solution is measured by using each of the following method:

1. **Henderson-Hasselbalch Equation:** use the equation to predict the pH of the buffer solution (compare this theoretical value with values obtained from experimental measurements). pKa for acetic acid is 4.76.
2. **Colorimetric Method:**
 - a) Paper indicator: immerse a strip of wide range pH paper into small quantity of buffer solution and observe the color changes of the paper.
 - b) Liquid universal indicator: add 2 drops of universal indicators to 10 ml buffer solution, then compare the color result with color found on the bottle of liquid universal indicator.
3. **pH Meter:** Calibrate the pH meter using the pH 4 buffer provided. Put the electrode of the pH meter in the buffer solution and read the pH.

3. Measurement of Buffer capacity:

1. Addition of Acid:

- Add small quantities (e.g., 5 mL, 10 mL, and 15 mL) of 0.1 M HCl to the acetate buffer solution.
- After each addition, stir the solution and measure the pH.
- Record the pH values after each addition.

2. Addition of Base:

- Similarly, add small quantities of 0.1 M NaOH (e.g., 5 mL, 10 mL, and 15 mL) to a fresh portion of the buffer solution.
- After each addition, stir the solution and measure the pH.
- Record the pH values after each addition of base.

3. Calculation of Buffer Capacity:

- Using the recorded data, calculate the buffer capacity (β) at each step using the formula:

$$\beta = \Delta B / \Delta pH$$

Example of calculation:

Calculation the buffer capacity of acetate buffer ($pH=4.76$) after addition of 10ml HCl

$$\beta = \Delta B / \Delta pH$$

- $\Delta pH = \text{Initial pH} - \text{Final pH after HCl added}$

Initial pH= 4.76

Final pH after HCl added (measured and recorded from the experiment)= 4.65

$$\Delta pH = 4.76 - 4.65 = 0.11 \quad (1)$$

- $\Delta B:$

$$0.1 \times 10 = C_2 \times 60$$

$$C_2 = 0.017$$

$$\Delta B = 0.017 \quad (2)$$

- $\beta = \Delta B / \Delta pH = 0.017 / 0.11 = 0.15$

Thus, the buffer capacity of our acetate buffer solution is **0.15**

- Notes:** A smaller change in pH (ΔpH) for a given ΔB indicates a higher buffer capacity.

Results

1. Results of addition the HCl to the acetate buffer solution:

NO.	pH before addition of 0.1 M HCl	Volume of 0.1 M HCl	pH after addition of 0.1 M HCl	Buffer capacity
1	4.76	5 ml		
2	4.76	10 ml		
3	4.76	15 ml		

Results

2. Results of addition the NaOH to the acetate buffer solution:

NO.	pH before addition of 0.1 M NaOH	Volume of 0.1 M NaOH	pH after addition of 0.1 M NaOH	Buffer capacity
1	4.76	5 ml		
2	4.76	10 ml		
3	4.76	15 ml		



Thank You