Experiment
Preparation of Buffer Solution and Measurement of pH

Aim

To prepare an acetate buffer solution and determine its pH using a pH meter, and to study its buffering action.

References

S.R. Kale; R.R. Kale: Practical Book of Biochemistry & Clinical Pathology; Nirali prakashan; 2020; 29th Edition Pahe no 12-14.

Requirements

- **Glassware Requirement:-** pH meter, Pipettes, beaker, volumetric flask, Glass rod.
- **Chemical Requirement:-** Acetic acid (0.1M), Sodium acetate (0.1M), Distilled water.

Principle

A buffer solution resists change in pH. It consists of a weak acid and its salt. The pH is given by Henderson-Hasselbalch equation: pH = pKa + log([Salt]/[Acid]).

For acetic acid: pKa=4.76

Procedure

A. Preparation of Buffer Solution

1. Pipette **50 ml of 0.1 M acetic acid** into a clean beaker.
2. Add **50 ml of 0.1 M sodium acetate**.
3. Mix thoroughly using a glass rod.
4. Transfer to a volumetric flask if required.

B. Calibration of pH Meter

1. Switch on the pH meter.
2. Calibrate using standard buffer solutions (pH 4.0, 7.0).
3. Rinse electrode with distilled water and blot dry.

C. Measurement of pH

1. Immerse the electrode into the prepared buffer solution.
2. Allow the reading to stabilize.
3. Record the pH value.
4. Repeat for 3 trials

D. Study of Buffer Action

1. Take 20 ml of prepared buffer in two separate beakers.
2. Add:
    - 1 ml of **0.1 M HCl** in one beaker
    - 1 ml of **0.1 M NaOH** in another
3. Mix and record the pH again.

Observations

1.Buffer Preparation

Solution	Volume (ml)	Concentration
Acetic acid	50 ml	0.1 M
Sodium acetate	50 ml	0.1 M

2.Theoretical pH Calculation

- pH=4.76+log(1)=4.76

  Theoretical pH = 4.76

3.Experimental pH Values

Trial No.	Observed pH
1	4.74
2	4.77
3	4.75

Average pH = 4.75

4.Buffer Action

Addition	pH Before	pH After
+1 ml 0.1 M HCl	4.75	4.62
+1 ml 0.1 M NaOH	4.75	4.88

Result

The prepared acetate buffer solution showed an experimental pH of **4.75**, which is very close to the theoretical value (**4.76**). The buffer resisted changes in pH on addition of acid and base, confirming its buffering capacity.