LABORATORY 3

BUFFER SOLUTIONS

Mixtures of a weak acid or a weak base, together with their hydrolysing salts, form a **buffer solution**. A buffer solution is resistant to pH change on addition of small amount of strong acid or strong base. In the presence of completely dissociated hydrolysing salt, the weak electrolyte (weak acid or weak base) does not dissociate. **Phosphate buffer** consists of two phosphate salts which introduce two phosphate ions to the solution: $\mathbf{H_2PO_4^-}$ and $\mathbf{HPO_4^{2-}}$**. These ions neutralize** both H^+ and OH^- ions added. After introduction of H^+ ions to the phosphate buffer, $\mathbf{HPO_4^{2-}}$ anions accept hydrogen ions (act as a Bronsted/Lowry base), forming $\mathbf{H_2PO_4^-}$ ions:

$$\mathbf{HPO_4^{2-} + H^+ \rightarrow H_2PO_4^- + H_2O}$$

As the hydrogen phosphate anion ($\mathbf{HPO_4^{2-}}$) concentration decreases, the dihydrogen phosphate anion ($\mathbf{H_2PO_4^-}$) concentration increases.

Introduction of $\mathbf{OH^-}$ ions to the buffer solution, causes the following reaction ($\mathbf{H_2PO_4^-}$ ions act as a Bronsted/Lowry acid because they donate H^+):

$$\mathbf{H_2PO_4^- + OH^- \rightarrow HPO_4^{2-} + H_2O}$$

As the concentration of dihydrogen phosphate anion ($\mathbf{H_2PO_4^-}$) decreases, the concentration of hydrogen phosphate anion ($\mathbf{HPO_4^{2-}}$) increases.

A **buffer's capacity (β)** to hold a constant pH depends <u>on the molar concentration of buffer components</u>. To calculate the buffer's capacity we can use equation:

$$\beta = \frac{\text{n - amount of acid or base added to buffer (mole/L)}}{\Delta \text{ pH}}$$

1. The influence of acid to salt molar ratio on pH of buffer

Procedure:

Prepare buffer's mixtures according to the table.

beaker number	0,1M **CH_3COOH** [cm^3]	0,1M **CH_3COONa** [cm^3]	H_2O [cm^3]	pH	$\frac{n_{acid}}{n_{salt}}$
1	20	10	10		
2	10	20	10		

- measure pH of prepared buffers
- calculate the ratio between acid and base in each solution
- take conclusions (explain how the change of the ratio between acid/salt in the buffer mixture influences pH)

2. The influence of buffer concentration on pH value and buffer capacity

Procedure:

Prepare buffer's mixtures according to the table.

beaker number	V [cm^3] **phosphate buffer**		V [cm^3] H_2O	pH_1	V [cm^3] added 0,05M HCl	pH_2	ΔpH	n_{HCl}	β
	0,1M	0,01M							
1	40	—	—		2				
2	—	40	—		2				
3	—	—	40		2				

- measure initial pH (pH_1) of the solutions and water
- add 2 ml of 0,05 M HCl to each beaker, mix and again measure pH (pH_2)
- calculate pH difference (ΔpH) and moles of acid (n) added to 1 liter of buffers or water
- calculate buffer capacity for buffers (not for water)
- draw the conclusions from the experiment (determine the relation between concentration of buffer and buffer capacity and compare pH changes in water and buffers occurring after addition of strong acid)

Problems

1. **1.** What is pH of the mixture composed of 20 g of NaOH (40 g/mol) and 1 liter of 2 M acetic acid? (K_a = $1.8 \cdot 10^{-5}$)? (Answer: $[H^+] = 5.4 \cdot 10^{-5}$)
2. **2.** Calculate pH of phosphate mixture composed of 0.01 mole KH_2PO_4 and 0.001 mole Na_2HPO_4 in 1 liter of the solution. $K_{H2PO4^-} = 2 \cdot 10^{-7}$. (Answer: pH = 5.7)
3. **3.** 1.2 mL of 0.1 M HCl was added to 10 mL of acetate buffer pH 5.4. pH decreased to 4.2. Calculate the buffer's capacity. (Answer: $\beta = 0.01$)
4. **4.** Enzymatic reaction was performed in 0.25 M phosphate buffer (0.125 mol/L $H_2PO_4^-$ and 0.125 mol/L HPO_4^{2-}), pH 6.8. As the result of the reaction 0.05 moles of H^+ were produced (per 1 L of the buffer). What was the final pH of the solution? What would be pH in case of a lack of a buffer ? $K_{H2PO4^-} = 1.58 \times 10^{-7}$. (Answer: $pH_1 = 6.43$, $pH_2 = 1.3$)
5. **5.** The concentration of H_2CO_3 in the human plasma is 1.25×10^{-3} mol/L. What is concentration of HCO_3^- if pH of the blood is 7.4? $K_{H2CO3} = 8 \times 10^{-7}$. (Answer: 25 mmol/L)
6. **6.** Bicarbonate buffer is the main buffer of human plasma responsible for regulation of blood pH. At pH 7.4, HCO_3^- ions concentration is 25 mM, CO_2 – 1.2 mM. What will be pH change after addition 0.005 moles of H^+ to 1 L of the blood? pK = 6.1. In the human body the excess of CO_2 is removed together with exhaled air and CO_2 concentration is not changed. (Answer: 7.3 is the final pH)
7. **7.** What is p_{CO2} in the patient blood if pH = 7.25, HCO_3^- = 25 mmol/L, $K_a = 8 \times 10^{-7}$, $\alpha = 0.03$ mmol/L · mmHg ($[CO_2] = \alpha \times p_{CO2}$)? What type of abnormality does the patient have? (Answer: 59 mmHg, acidosis)
8. **8.** How many mL of 2 % (m/v) $NaHCO_3$ (84 g/mol) should be given to patient to increase pH of the blood from 7.05 to 7.45 ? P_{CO2} = 35 mmHg; V_{plasma} = 3 L. (Answer: 178.2 mL)