Buffer System

- **Acid** is a substance that can **release hydrogen ions** (protons H^+).
- **Base** is a substance that can **accept hydrogen ions.**
- **pH** is the concentration of hydrogen ions it determines the acidity of the solution
- The pH of a solution is the negative base 10 logarithm of its hydrogen ion concentration
- $\mathbf{pH} = -\log_{10}[H^+]$

- The “p” in pH or in pKa signifies -log

pH	$[H^+]$	
0	(10^{0})	1.0
1	(10^{-1})	0.1
2	(10^{-2})	0.01
3	(10^{-3})	0.001
4	(10^{-4})	0.0001
5	(10^{-5})	0.00001
6	(10^{-6})	0.000001
7	$(\mathbf{10^{-7}})$	**0.0000001**
8	(10^{-8})	0.00000001
9	(10^{-9})	0.000000001
10	(10^{-10})	0.0000000001
11	(10^{-11})	0.00000000001
12	(10^{-12})	0.000000000001
13	(10^{-13})	0.0000000000001
14	(10^{-14})	0.00000000000001

pH	$[H^+]$ (mol/l)	
1	10^{-1}	↑
2	10^{-2}	
3	10^{-3}	Increasing acidity
4	10^{-4}	
5	10^{-5}	
6	10^{-6}	
7	10^{-7}	Neutral
8	10^{-8}	
9	10^{-9}	
10	10^{-10}	Increasing alkalinity
11	10^{-11}	
12	10^{-12}	
13	10^{-13}	
14	10^{-14}	↓

- The following examples illustrate how to calculate the pH of acidic and basic solutions.
- ***Example 1:*** What is the pH of a solution whose hydrogen ion concentration is 3.2×10^{-4} mol/L?
- $pH = -\log [H^+]$

  $= -\log (3.2 \times 10^{-4})$
- $= -\log (3.2) - \log(10^{-4})$
- $= -0.5 + 4$
- $pH = 3.5$

- When an acid loses a proton, its conjugate base is formed.
- The tendency of any acid (HA) to lose a proton and form its conjugate base (A-) is called **dissociation constants (Ka) and thus measure the strength of an acid.**

$$\underset{\text{Acid}}{HA} \rightleftharpoons \underset{\text{Conjugate Base}}{A^-} + H^+$$

$$K_a = \frac{[A^-]\,[H^+]}{[HA]}$$

- The stronger the acid, the greater its tendency to lose its proton.
- **<u>Strong acids</u>**: are acids that dissociate completely in solution like HCl. $HCl \rightarrow Cl^- + H^+$
- **<u>Weak acids</u>**: are acids that dissociate only to a limited extent like H_2CO_3. $H_2CO_3 \rightarrow HCO_3^- + H^+$
- The weak **acid** (proton donor) dissociates into a hydrogen ion H+ and an anionic component (A-), called the **conjugate base** (or salt).

pKa

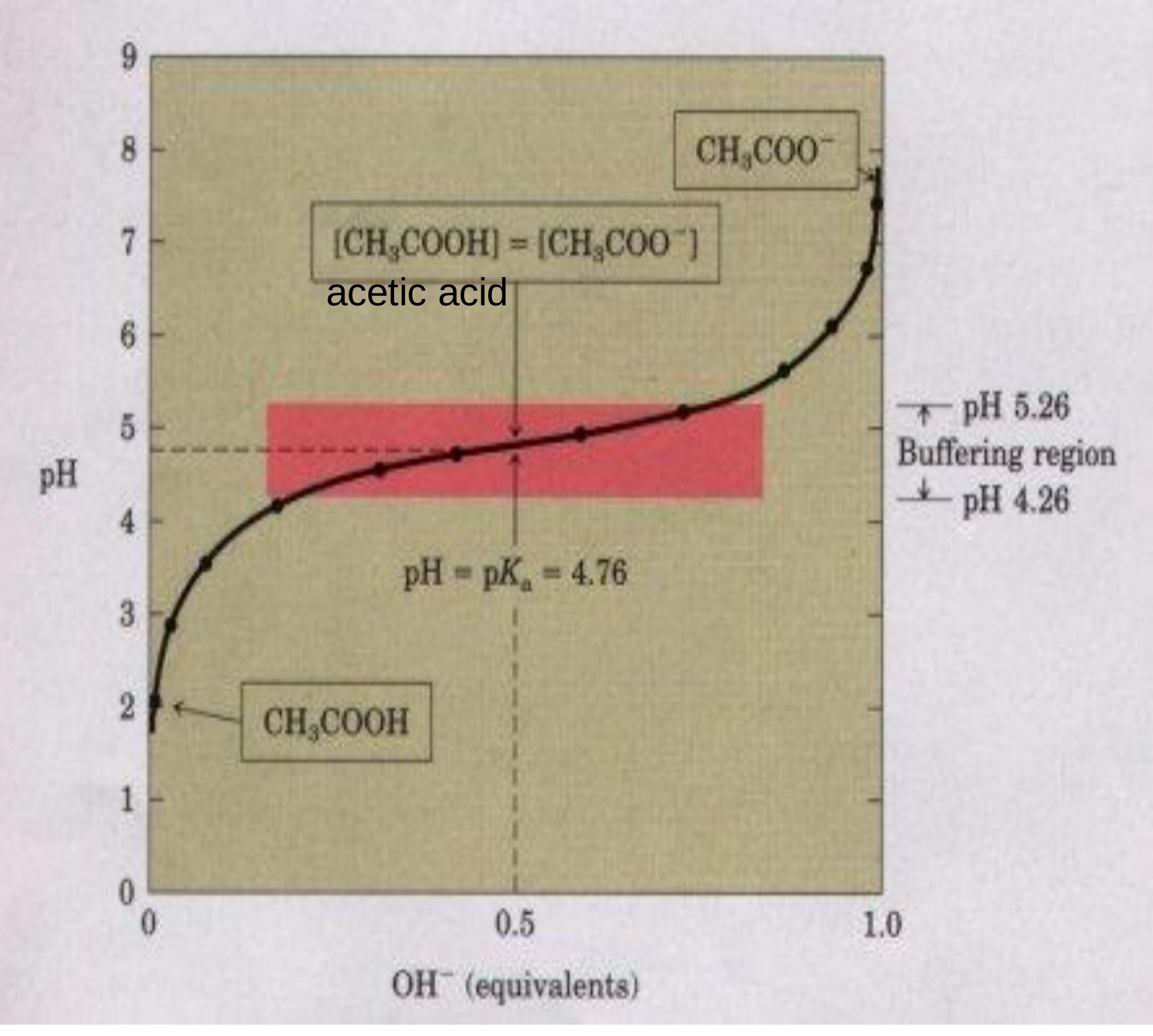
- pKa = -log Ka $\quad$ **Ka = 10$^{(-pKa)}$**

- **pKa of an acid is the pH at which 50% dissociation occurs**
- pKa value is easier to work with and remember than Ka value as of H and pH.

- Strong acids has strong tendency to dissociate and thus has high Ka value and low p*K*a value and thus the lower the pH the compound will produce in solution.

- Example a <u>strong acid</u> with Ka of 10^7 has a pKa of -7, while a <u>weak acid</u> with Ka of 10^{-12} has a pKa of 12

The maximum buffering capacity exists when the pH of the solution equals the pK' of the buffer, [conjugate base] = [acid], the buffer can then respond equally to both added acid and added base



Buffers

- A buffer is a solution that <u>resists</u> pH changes when acids or bases are added to the solution.
- Buffer solutions consist of a weak acid (undissociated acid) and its conjugate base (the form of the acid having lost its proton).

$$\underset{\text{Acid}}{HA} \rightleftharpoons \underset{\text{Conjugate Base}}{A^-} + H^+$$

- A buffer works because added acids (H^+) are neutralized by the conjugate base (A^-) which is converted to the acid (HA).
- Added bases are neutralized by the acid (HA), which is converted to the conjugate base (A^-).
- <u>Two factors determine the effectiveness of a buffer</u>:

1- its pKa relative to the pH of the solution

2- its concentration.

Henderson-Hasselbalch Equation

- Henderson-Hasselbalch adjusted equation shown below describe the relationship between the acid and its conjugate base with pH and pKa

$$\boxed{pH = pKa + \log \frac{A^{-}}{HA}}$$

$$\mathbf{pH = pK_a + \log_{10} \frac{[Conjugate\ Base]}{[Acid]}}$$

- The most effective buffers is when pH=pKa means it has equal concentrations of acid [HA] and its conjugate base [A-] **(**50% of both forms AH & A- present in solution).
- At pH = p*K*a ± 1 the buffer capacity falls to 33% of the maximum value. Therefore the buffer is effective one point up or down the pH pKa value.

Solving Problems Using the Henderson-Hasselbalch Equation

1. Calculate the pK_a of lactic acid, given that when the concentration of lactic acid is 0.010 M and the concentration of lactate is 0.087 M, the pH is 4.80.

$$\text{pH} = \text{p}K_a + \log \frac{[\text{lactate}]}{[\text{lactic acid}]}$$

$$\text{p}K_a = \text{pH} - \log \frac{[\text{lactate}]}{[\text{lactic acid}]}$$

$$= 4.80 - \log \frac{0.087}{0.010} = 4.80 - \log 8.7$$

$$= 4.80 - 0.94 = 3.9 \quad (\textit{answer})$$

2. Calculate the pH of a mixture of 0.10 M acetic acid and 0.20 M sodium acetate. The pK_a of acetic acid is 4.76.

$$\text{pH} = \text{p}K_a + \log \frac{[\text{acetate}]}{[\text{acetic acid}]}$$

$$= 4.76 + \log \frac{0.20}{0.10} = 4.76 + 0.30$$

$$= 5.1 \quad (\textit{answer})$$

Organs controlling pH

1. Lungs function to regulate blood pH through bicarbonate system. The respiratory tract can adjust the blood pH upward in minutes by exhaling CO_2 from the body.

2. Kidney maintain a normal pH through:

A. Reabsorption of filtered bicarbonate.

B. Excretion of acids.

- The renal system can also adjust blood pH but this process takes hours to days to have an effect.

Acids in our Body

1. Volatile acid: represented in our body by carbonic acid which is originated from CO_2. So the main source of volatile acid is CO_2 which can evaporate and get rid of it through lungs.

2. Nonvolatile acids: include all acids produced in the body except the one that is produced from CO2 example lactic acid (fermentation), phosphoric acid, sulfuric acid (Protein breakdown), acetoacetic acid and *beta*-hydroxybutyric acid (ketone bodies).

- Nonvolatile acids elimination is through the kidney.

<u>Transport of CO_2</u>

- **<u>CO_2 is carried in the blood by 3 ways</u>:**

1- About 10% of the CO_2 in blood is simply dissolved in plasma.

2- About 20% of CO_2 react <u>nonenzymaticaly</u> with amino groups (NH_2 terminal amino group) of hemoglobin to form **carbamino hemoglobin (carbamate)**

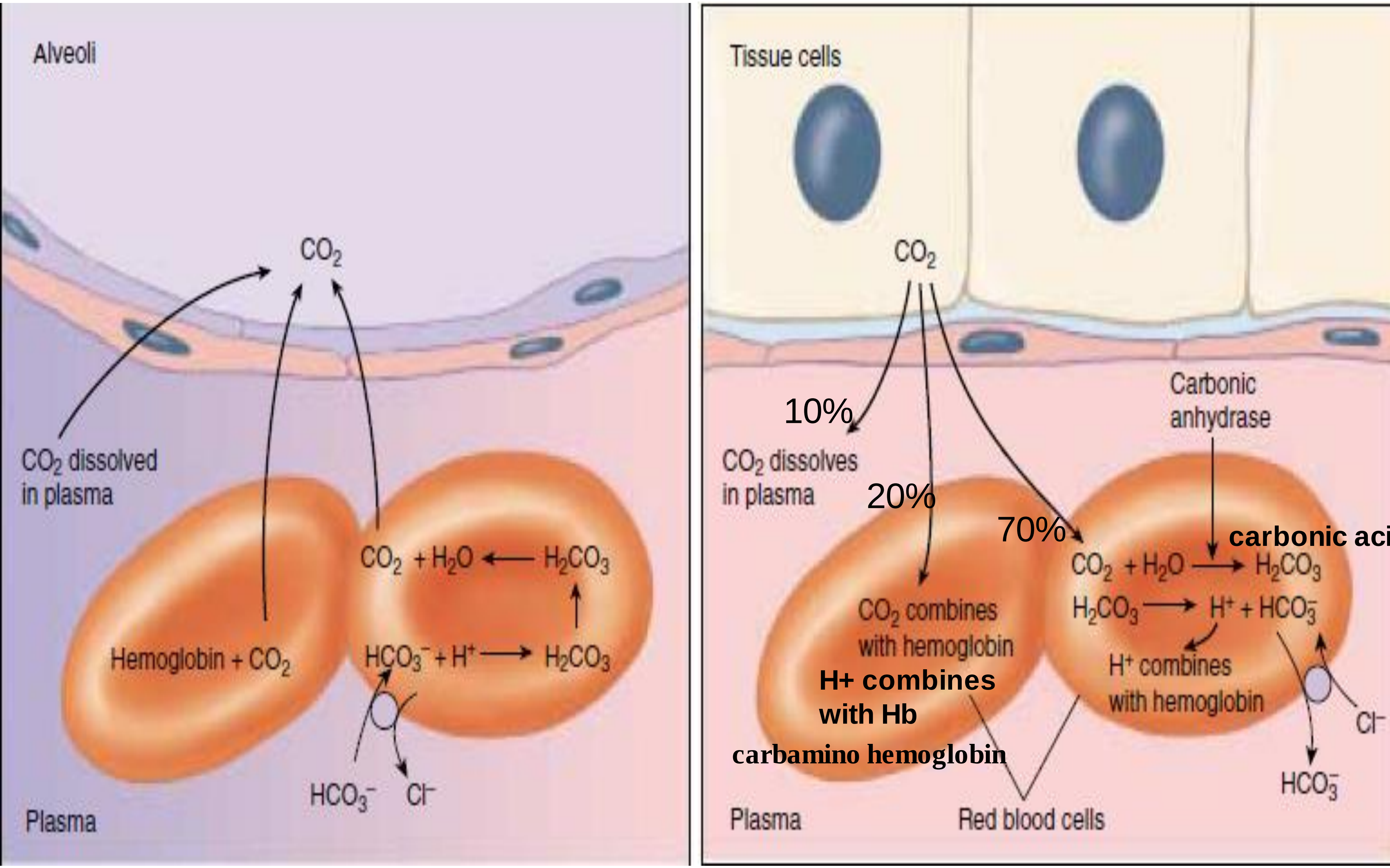
- $$\mathbf{HbNH_2 + CO_2 \leftrightarrow HbNHCOO^- + H^+}$$
- The excess H^+ produced binds with Hb and stabilize the deoxy form and promoting the release of O_2 to cells.

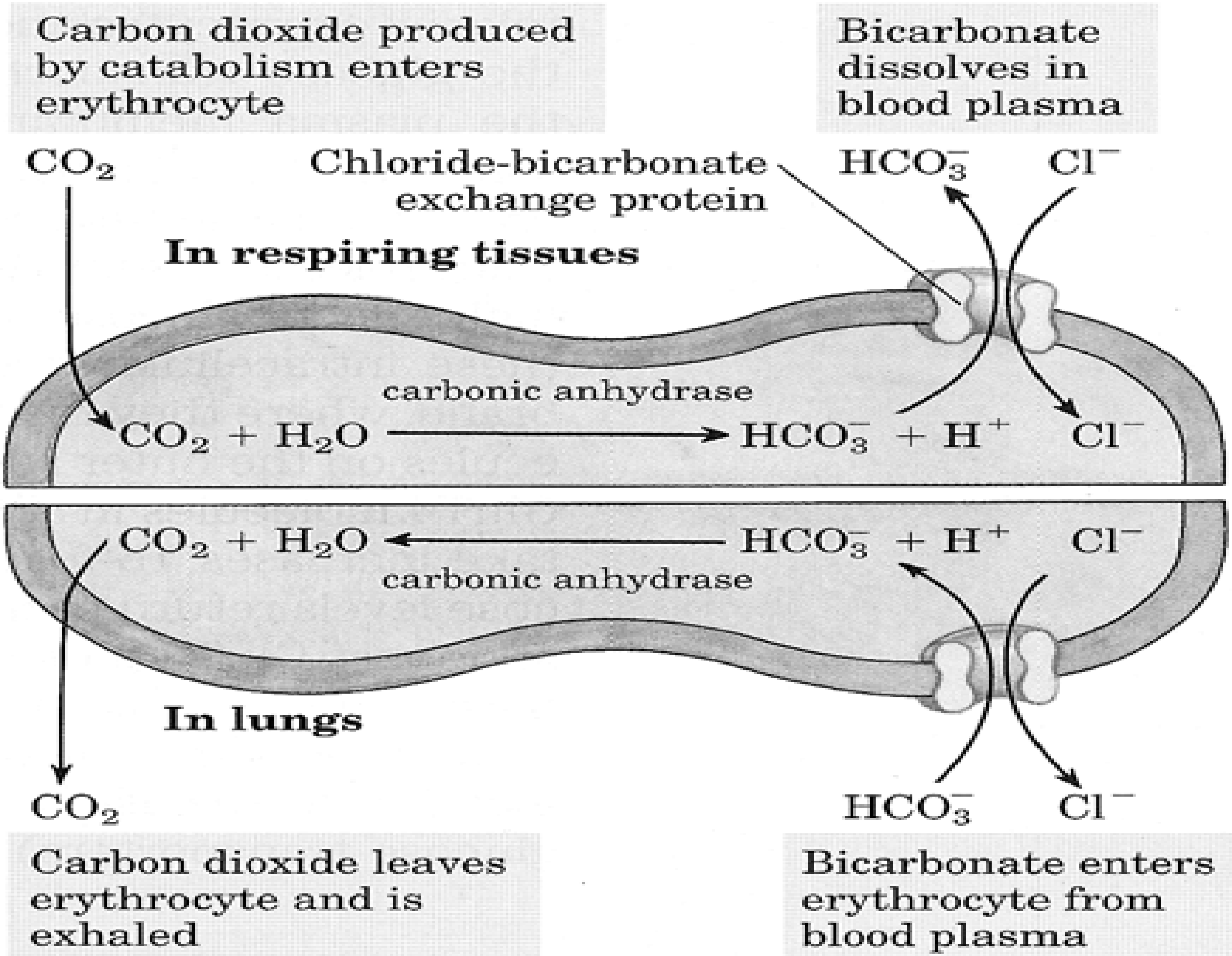
- In the lungs the high pO_2 concentration generates $Hb(O_2)_4$ with dissociation of H+. The increase in H+ forces dissociation of the carbamino group with release of CO2 which is expired from lungs.

3- The remaining 70% of the CO_2 diffuses into the red blood cells, where the enzyme carbonic anhydrase catalyzes the combination of CO_2 with water (hydration reaction) to form carbonic acid (H_2CO_3).

- Carbonic acid dissociates into bicarbonate (HCO_3–) and hydrogen (H+) ions. The H+ binds to hemoglobin and force $Hb(O_2)_4$ to dissociate its O_2 which diffuses out of RBC. While the bicarbonate moves out of the erythrocyte into the plasma via a transporter that exchanges one chloride ion for a bicarbonate (this is called the "chloride shift").
- The blood carries bicarbonate to the lungs. The lower pCO_2 of the air inside the alveoli causes the carbonic anhydrase reaction to proceed in the reverse direction that leads to formation of CO_2. The CO_2 diffuses out of the red blood cells and into the alveoli, so that it can leave the body in the next exhalation.

Transport of CO_2 by the blood




Carbon dioxide produced by catabolism enters erythrocyte
Bicarbonate dissolves in blood plasma
CO_2
Chloride-bicarbonate exchange protein
HCO_3^-
Cl^-
In respiring tissues
carbonic anhydrase
$CO_2 + H_2O \rightarrow HCO_3^- + H^+$
Cl^-
$CO_2 + H_2O \leftarrow HCO_3^- + H^+$
Cl^-
carbonic anhydrase
In lungs
CO_2
HCO_3^-
Cl^-
Carbon dioxide leaves erythrocyte and is exhaled
Bicarbonate enters erythrocyte from blood plasma

The Bicarbonate Buffer System

- Bicarbonate and other buffers normally maintain the pH of extracellular fluid in human's body between 7.35 and 7.45.
- The major source of metabolic acid in the body is the gas $CO_{2,}$ produced principally from fuel oxidation in the TCA cycle.

$$\mathbf{CO_2 + H_2O \;{}^{\text{carbonic anhydrase}}\; H_2CO_3 \qquad\qquad HCO_3^- + H^+}$$

- The pH of a bicarbonate buffer system depends on the concentration of H_2CO_3 as proton donor and HCO_3^- as proton acceptor.
- The pH of a bicarbonate buffer exposed to a gas phase is ultimately determined by the concentration of HCO_3^- in the aqueous phase and the partial pressure of CO_2 in the gas phase.

$$CO_2 + H_2O \rightleftharpoons H_2CO_3 \rightleftharpoons HCO_3^- + H^+$$

When acid is added:

$$HCO_3^- + H^+ \rightleftharpoons H_2CO_3$$

When a base is added:

$$H_2CO_3 + OH^- \rightleftharpoons HCO_3^- + H_2O$$

- **The Buffer Equation(s)**: The Henderson-Hasselbalch equation, is important for understanding buffer action in the blood and tissues
- **For bicarbonate system**: the normal average level of plasma bicarbonate of plasma is 24 mmol/litre. The normal CO_2 dissolved concentration in blood is 1.2 mmol/L (therefore the ratio of HCO_3^- to H_2CO_3 at pH 7.4 is 20 to 1 because most of the body's metabolic wastes, such as lactic acid and ketones, are acids). The pKa for carbonic acid is 6.1:
- pH = 6.1 + log 24/1.2
- = 6.1 + log20
- = 6.1 + 1.3
- = 7.4 which is the normal pH of arterial blood.

$$pH = pKa + \log \frac{[HCO_3^-]}{Pco_2}$$

- **What happen when CO_2 is increased**? Suppose that the CO_2 concentration doubled from 1.2 to 2.4. The doubling of CO_2 is achieved by increasing the PCO_2 in the atmosphere. Thus calculating pH from the above equation = **7.1**
- **What happen when increasing the concentration of HCO_3^-.** Increasing the conc of HCO_3^- from 24 to 48 will cause a change in pH from 7.4 to **7.7**
- Thus, removing CO_2 through lungs from the blood helps increase the pH and removing HCO_3^- from the blood helps lower the pH.

- The respiratory center in brain which controls the rate of breathing, is sensitive to changes in pH.
- As the pH falls, individuals breathe more rapidly and expire more CO_2.
- As the pH rises, they breathe more slowly.
- Thus, the rate of breathing contributes to regulation of pH through its effects on the dissolved CO_2 content of the blood.

Hemoglobin as Protein Buffer

- Hemoglobin in blood is made up of 574 amino acid, 36 of them are histidine
- The pKa of the various histidine residue (imidazole groups) on the different plasma proteins range from about 5.5 to about 8.5 thus providing a broad spectrum of buffer pairs.

- As stated before, the carbonic acid dissociates into bicarbonate anion and H+.
- The H+ released bind the side chain of the amino acid histidine (His-146 (β)) in the two β chains of hemoglobin.
- Only a small number of hydrogen ions generated in the blood remains free not attached to Hb. This explains why the acidity of venous blood (pH = 7.35) is only slightly greater than that of arterial blood (pH = 7.45).

Phosphate buffer the Intracellular pH

- Phosphoric acid (H_3PO_4) dissociate to conjugate base dihydrogen phosphate ion ($H_2PO^-{}_4$) and H+
- Dihydrogen phosphate ion dissociate to conjugate base hydrogen phosphate ($HPO^{2-}{}_4$) and H+ with a pKa of 7.2 which is very close to physiologcal pH
- **$H_2PO_4^- \leftrightarrow HPO_4^{-2} + H^+$**
- Thus, phosphate anions play a major role as an intracellular buffer in the red blood cell and in other types of cells, where their concentration is much higher than in blood and interstitial fluid.
- Organic phosphate anions, such as glucose 6-phosphate and ATP, also act as buffers.

- **Respiratory acidosis**
- Blood pH reflects changes in pH in tissues and values above or below the normal range 7.35—7.45 indicates a potential pathological condition. Blood pH below 7 or above 7.8 are life threatening and medical intervention is necessary. If the blood pH falls below 7.35 the condition is referred to as an acidosis and above 7.45 as alkalosis.
- Conditions of acidosis and alkalosis are divided according to the source into metabolic or respiratory.
- Respiratory acidosis is caused by hypoventilation so there is retention of CO_2 and a drop in pH thus the concentration of dissolved CO_2 in the blood increases, making the blood too acidic and is caused by condition restricting the exhaling of CO_2 from the lungs such as
- Diseases of the airways (such as asthma and chronic obstructive lung disease)
- Diseases of the chest (such as sarcoidosis)
- Diseases affecting the nerves and muscles that "signal" the lungs to inflate or deflate
- Depression of the respiratory centres in the medulla by different drugs.
- Severe obesity, which restricts how much the lungs can expand

- **Respiratory alkalosis**
- Results from hyperventilation that causes too much dissolved CO_2 to be removed from the blood, which decreases the carbonic acid concentration, which raises the blood pH. Often, the body of a hyperventilating person will react by fainting, which slows the breathing.
- Respiratory alkalosis may be caused from hysteria (any psychological dysfunction of unknown cause), central nervous system diseases, overdose of some drugs (e.g salicylate) and fever.