TAYLOR'S UNI. COLLEGE
CHEMISTRY (9701)
A Level

APPLICATION CHEMISTRY:
BIOCHEMISTRY
(Part 2 - Enzymes)

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(Yellow Room; Table 1)

Enzymes

Table 2: Comparison of the catalytic efficiency of certain enzymes

enzyme	turnover number (molecules reacted per enzyme molecule per minute)
carbonic anhydrase	36 000 000
catalase	5 600 000
β-amylase	1 100 000
β-galactosidase	12 500
phosphoglucose isomerase	1 240
succinate dehydrogenase	1 150

- Enzymes are **very specific**: catalysing only one particular reaction.
- **Each enzyme has a specific substrate**, the substrate being the target molecule acted upon during the enzyme-catalysed reaction.

Enzymes

- **Higher reaction rates –** the rates increased by factors of 106 to 1012 times compared to the uncatalysed reaction and are several orders of magnitude greater than those of the corresponding chemically catalysed reaction.

- **Milder conditions –**temperatures below 100°C, atmospheric pressure, and at pH's around neutrality.

- **Greater reaction specificity –** enzymes have a vastly more defined specificity with regard to their substrate and products: enzyme catalysed reactions are 'clean' and do not produce side products.

- **Capacity for regulation –** the catalytic activities of many enzymes can be varied by the concentrations of substances other than the substrate: the mechanism of these regulatory processes can be complex.

Shape of Enzymes

- **Water-soluble globular** proteins.
- **Complicated folding** of the protein chain to form the **tertiary structure** gives rise to **‘clefts’** or **‘crevices’**
- Precise shape of these clefts is designed to **‘recognise’ and hold in place a particular substrate molecule** while it reacts.
- Known as the **active site** of the enzyme.

Shape of Enzymes

- Catalytic **properties** and **specificity** of an enzyme are determined by :

a) the **shape** that matches the substrate

b) chemical nature of the amino acid **R-groups** located at the **active site**.

‘Lock and Key’ Model

- Only one substrate will fit into the active site, just as only one key fits a lock.

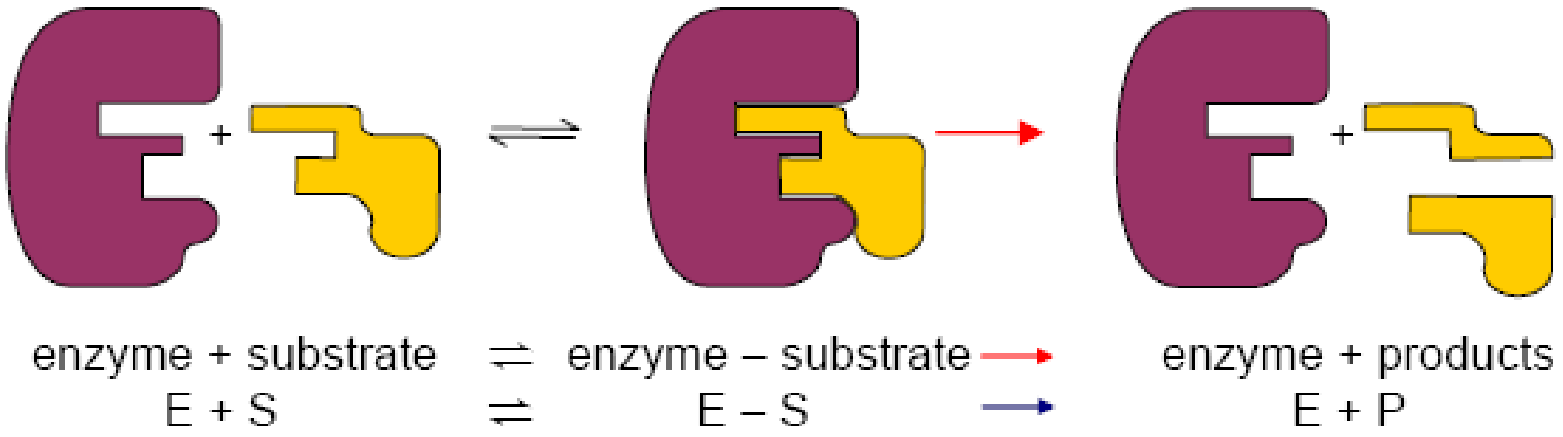


Figure 1.18 – the ‘lock and key’ mechanism

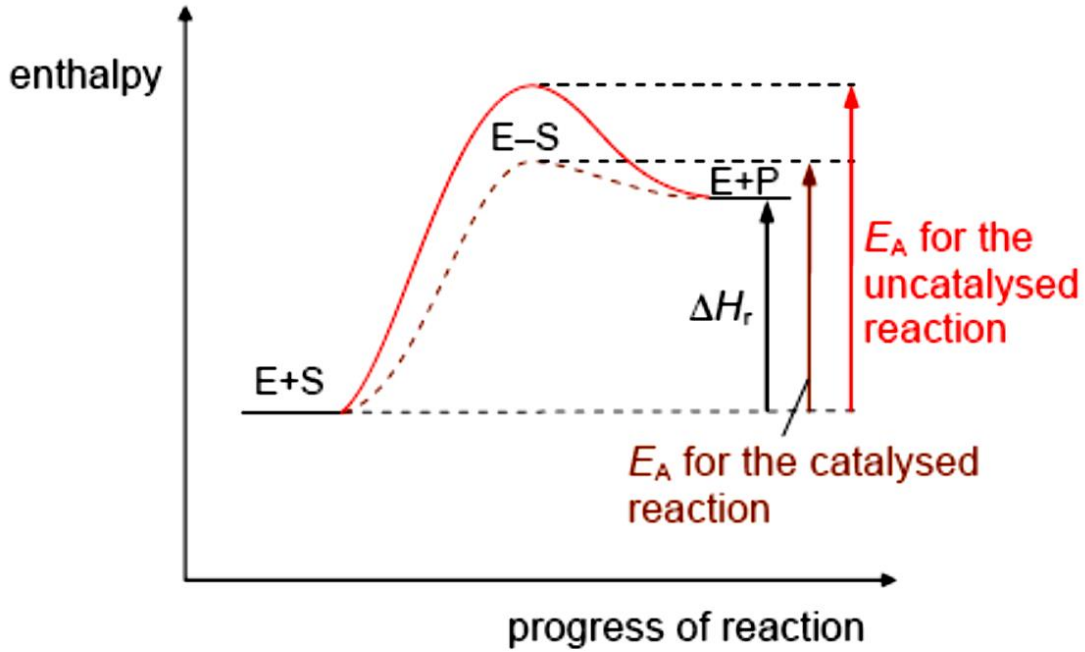


Figure 1.19 – energy profile for enzyme-catalysed and non-catalysed reaction

- **First stage of the reaction is reversible** since if the available energy is not greater than E_a the complex may dissociate without product being formed.

- In some cases the second stage is also reversible, making the whole enzyme catalysed process capable of proceeding in either direction depending on the cells metabolic requirements.

Competitive Inhibition

- **competitive inhibitor → imitates the substrate in the way it binds to the enzyme**.
- have a **similar shape to the substrate molecule** - **can bind to the active site** but **cannot participate in the catalysed reaction**.
- When they are present in the active site no reaction is taking place and the correct substrate cannot attach to the enzyme.

Competitive Inhibition

- Competition between the substrate and the inhibitor to occupy the active sites on the enzyme molecules.
- Result of this **competition depends on the relative concentrations** of the substrate and inhibitor.
- Functionality of the enzyme molecules is not interfered with – the active sites are merely blocked (Figure 1.21).
- This type of **inhibition is reversible by an increase in substrate concentration**.
- The max. rate of reaction is the same both with or without competitive inhibitor.

Competitive Inhibition

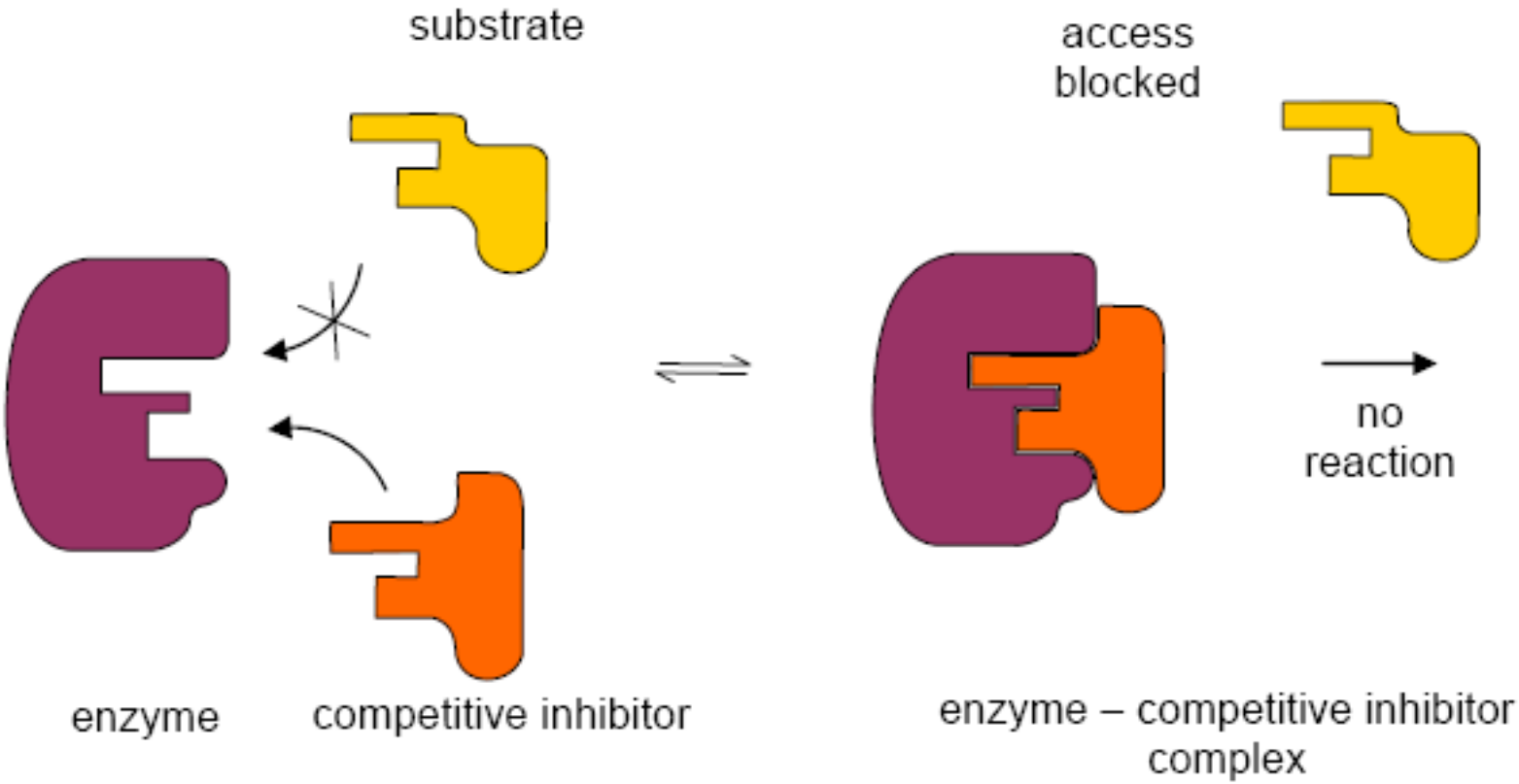


Figure 1.21 – model of action of a competitive inhibitor

Competitive Inhibition

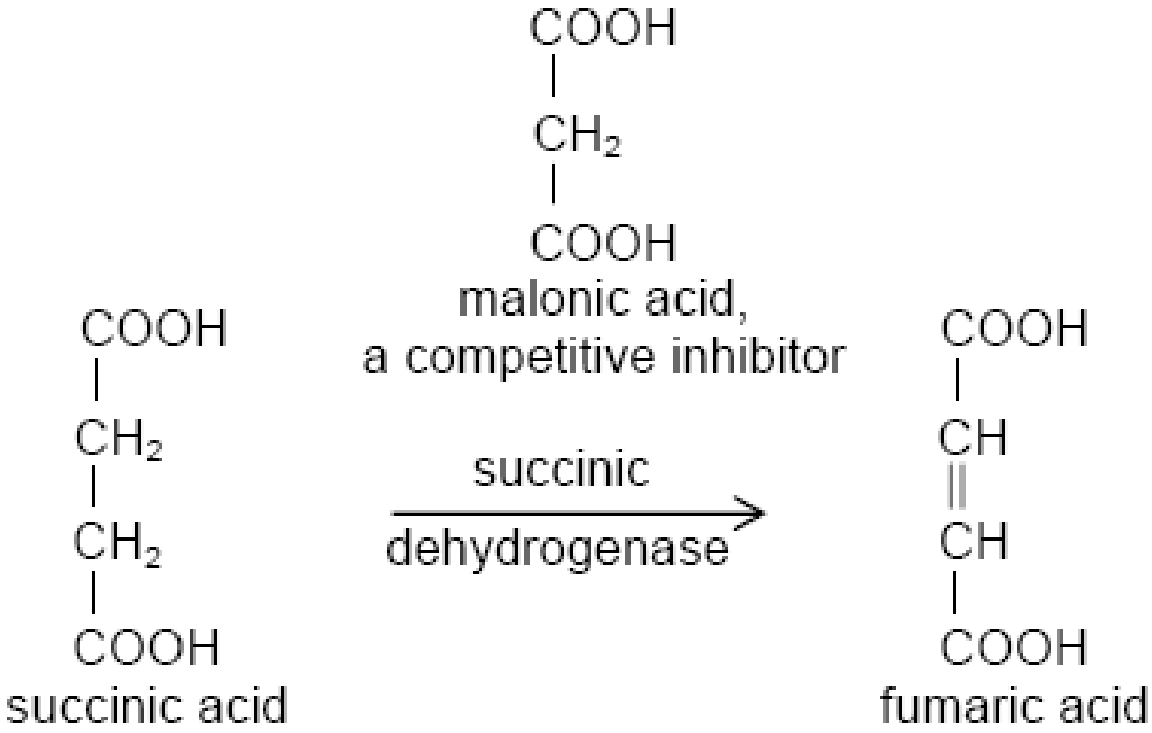


Figure 1.22 – succinate, the reaction, and some competitive inhibitors

Non-competitive Inhibition

- Molecules **can bind on to regions of the enzyme other than the active site** and affect enzyme activity.

- This binding is thought to cause one of the following:

  – the **active site to change shape** so that the **substrate cannot bind** (Figure 1.23).

  – the **enzyme-substrate complex to change shape so that the reaction cannot take place**.

Non-competitive Inhibition

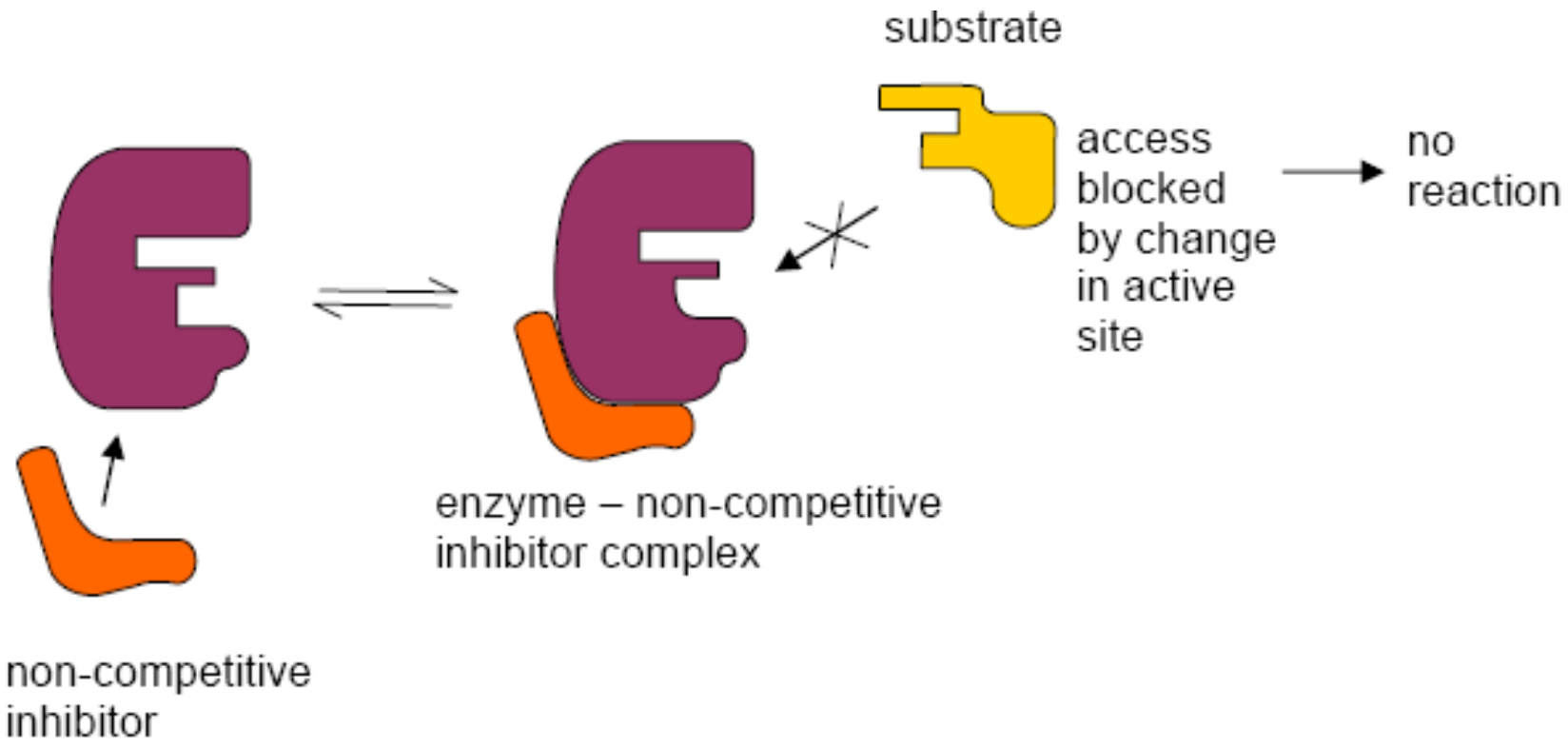


Figure 1.23 – scheme for non-competitive inhibition

Non-competitive Inhibition

- This inhibition **cannot be overcome simply by adding more substrate**.

- Effect - reduce the number of active enzyme molecules available for reaction.
- Hence max. rate of reaction is lowered.

- Most non-competitive inhibitors only bind weakly to the enzyme.

- This type of inhibition is **reversible** if the concentration of inhibitor falls.
- →the enzyme-inhibitor complex falls apart and the functional shape of the enzyme is restored.

Non-competitive Inhibition

- E.g : Heavy metal ion (Ag or Hg)
- The heavy metal ions react reversibly with one or more –SH groups, replacing the hydrogen atom with a heavy metal atom

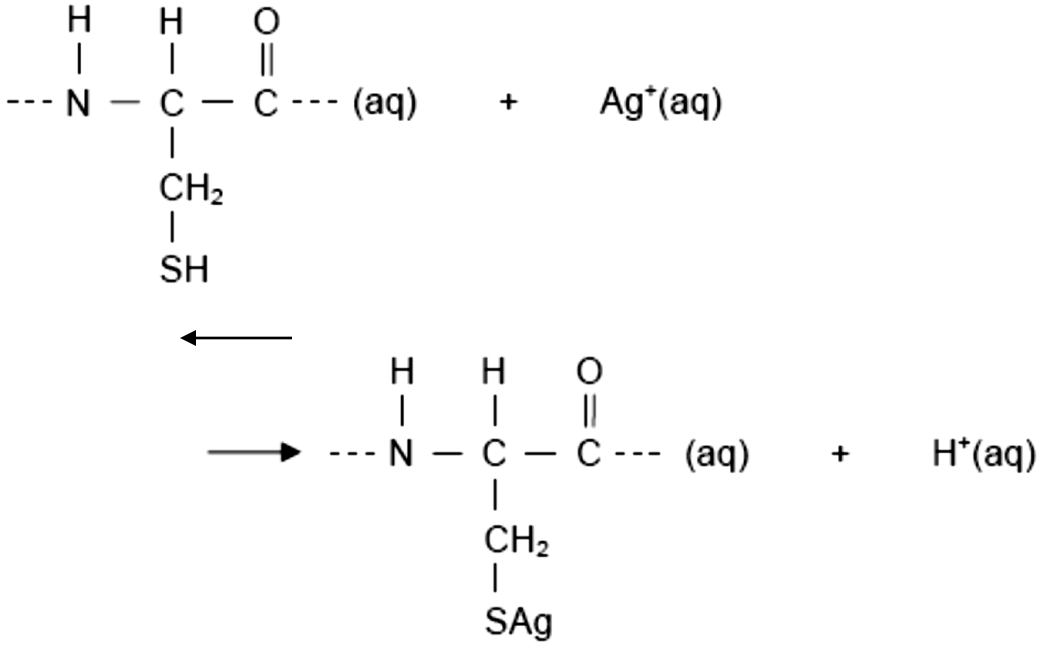
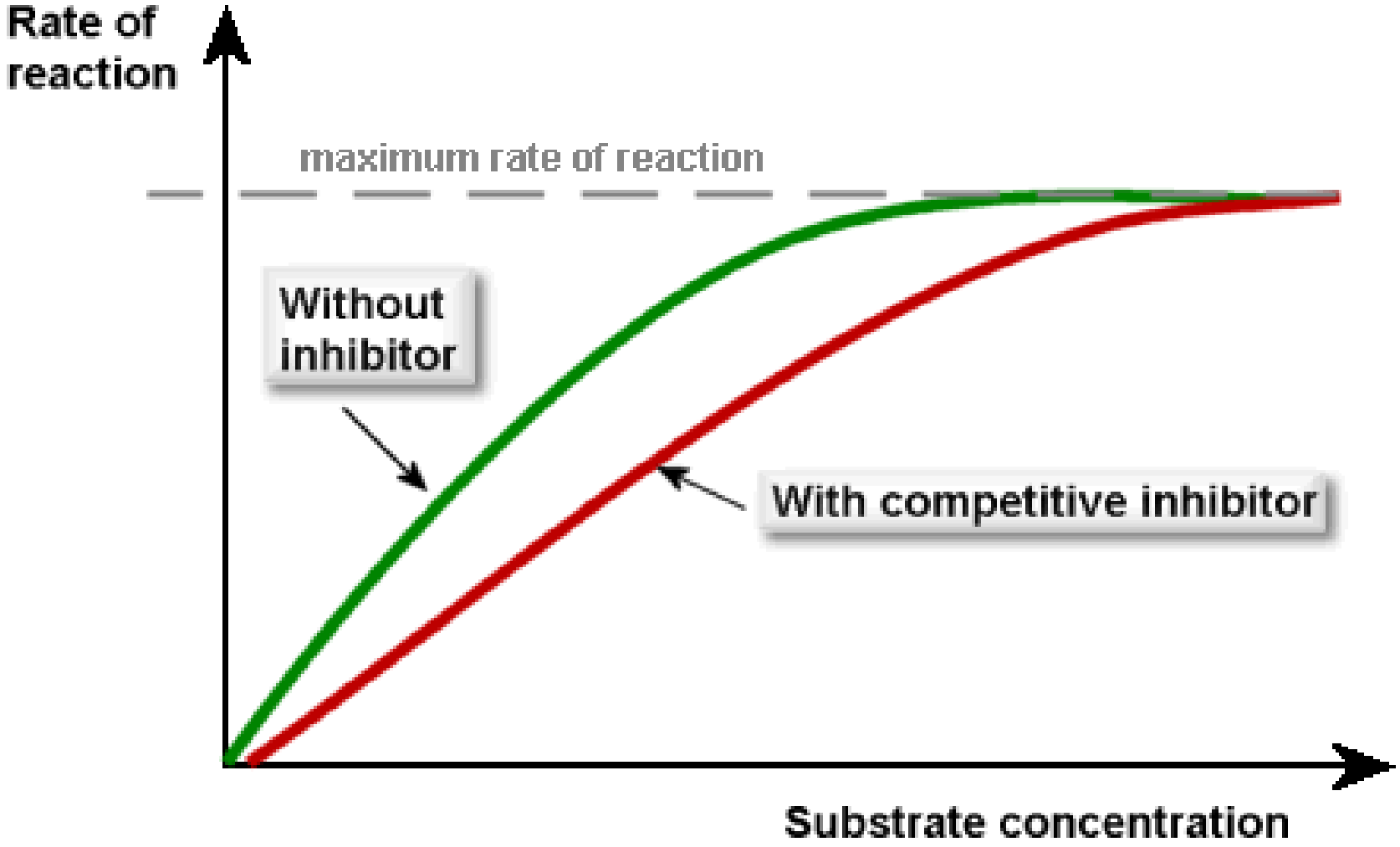


Figure 1.24 – the reaction between heavy metal ions and an –SH group



http://www.youtube.com/watch?v=PILzvT3spCQ&feature=related

Factors Affecting Enzyme Activity

Temperature

- Effect of temperature on enzyme activity is complex because it is the outcome of several different factors:
  - the **speed of the molecules**;
  - the **activation energy** of the catalysed reaction;
  - the **thermal stability of the enzyme and the substrate**.

Factors Affecting Enzyme Activity

Temperature

- At relatively low temperatures (around 0°C) the rate of most enzyme-catalysed reactions is very low – enzyme deactivated.

- Increasing temperature increases the rate of enzyme activity. Between 0°C and approximately 40°C the rate of enzyme activity increases almost linearly.
- Why?

Factors Affecting Enzyme Activity

Temperature

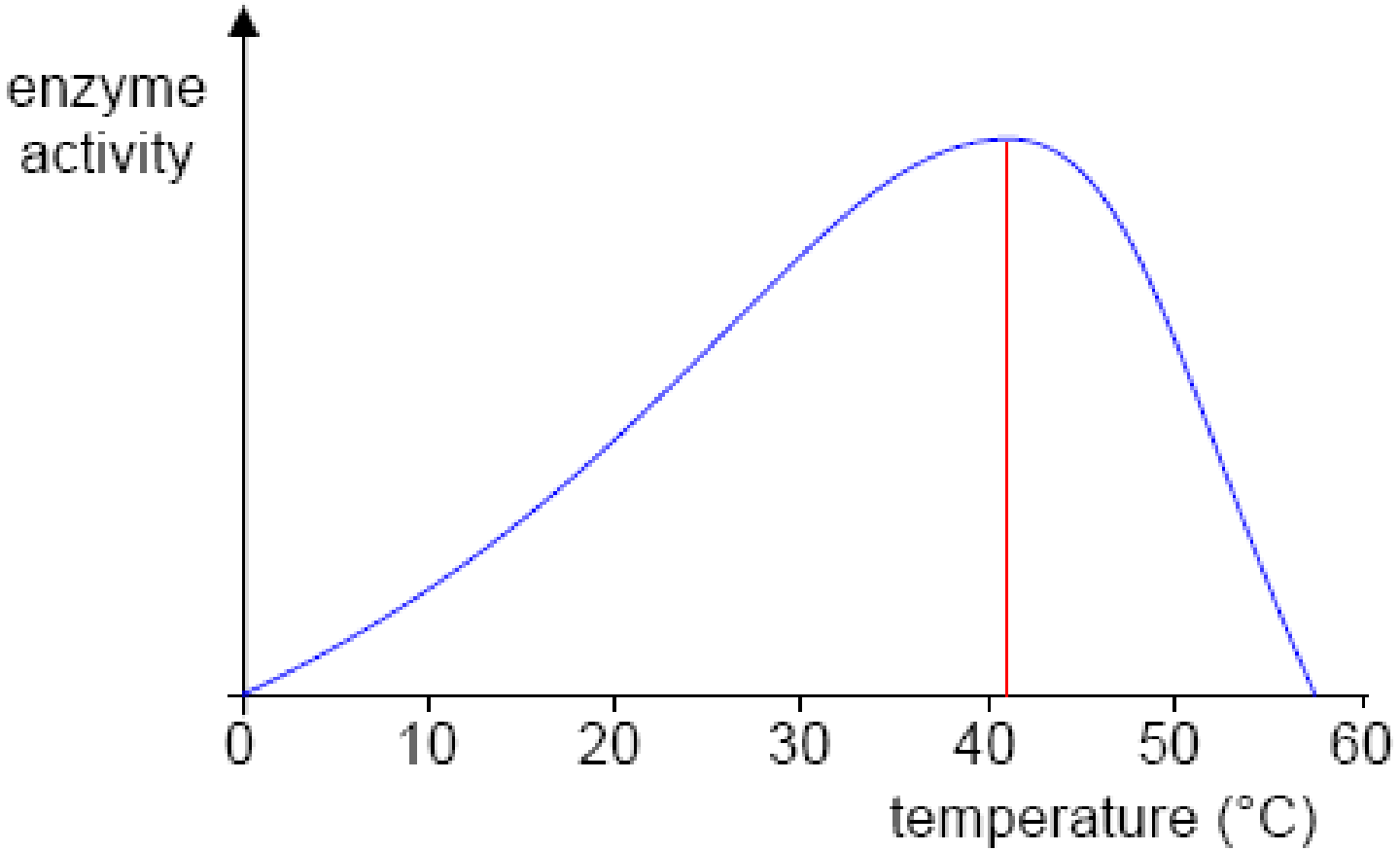


Figure 1.25 – profile of enzyme activity with temperature

Factors Affecting Enzyme Activity

Temperature

- Rate of reaction starts to decrease above 40°C.
- **Increased thermal motion** of the polypeptide chain is causing disruption of the forces maintaining the shape of the enzyme molecules.
- The enzyme molecules are progressively **denatured**, causing the **shape** of the **active site** to **change**.
- **Above 65°C** the enzymes from most organisms are **completely heat denatured**.

Factors Affecting Enzyme Activity

pH

- **Extreme pH** (high acidity or alkalinity) **will denature proteins** by **disrupting the precise three-dimensional arrangement of the protein chains**.

- If **enzyme activity depends on** particular residues in the **active site being charged or not**, then a **shift of just one pH unit can change the enzyme activity** significantly.

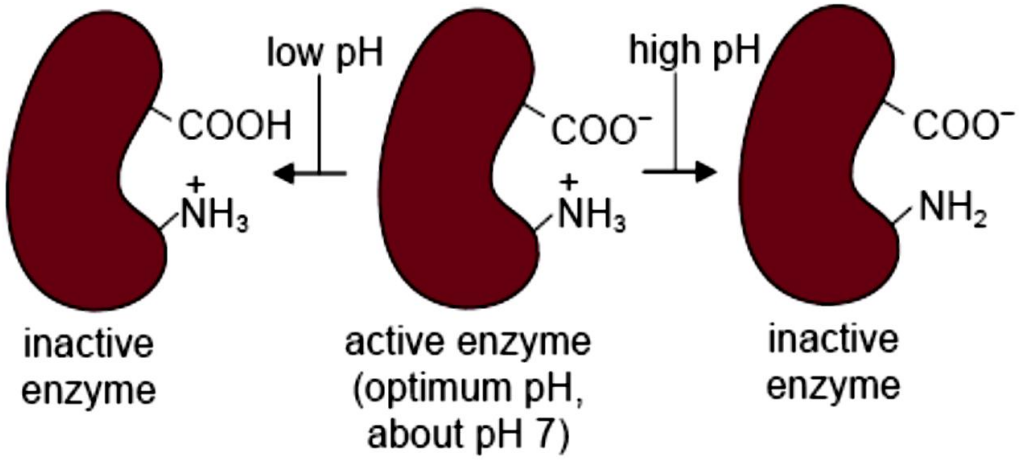


Figure 1.26 – pH changes can affect the ionization of the active site

Factors Affecting Enzyme Activity

pH

- Most enzymes are active over a fairly narrow range of pH.
- Each enzyme has its own distinct optimum pH (Figure 1.27).

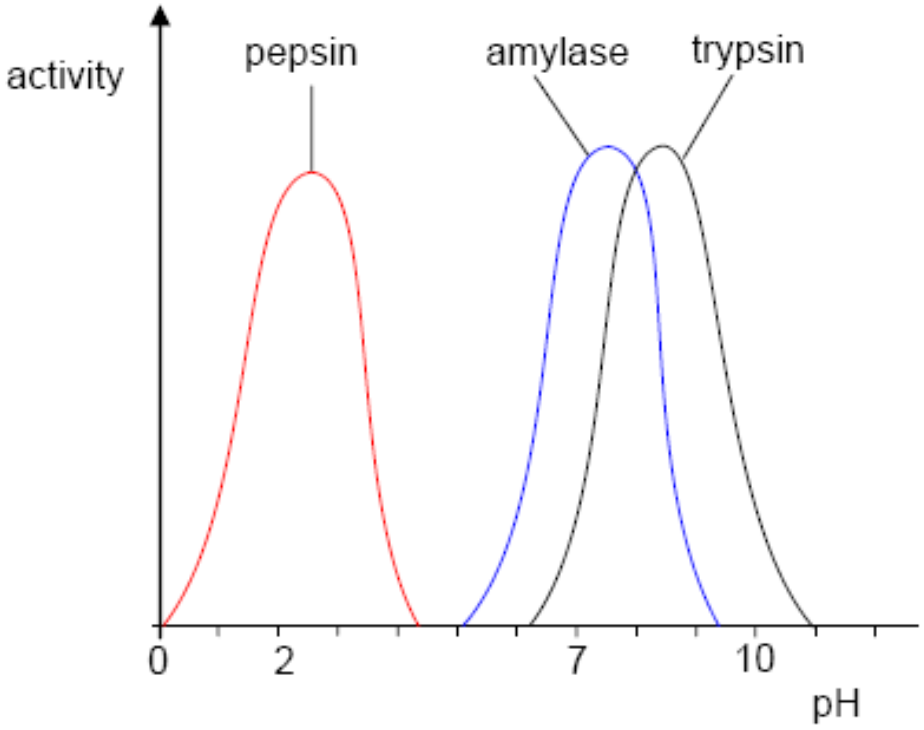


Figure 1.27 - curves showing pH optima for several enzymes

Chemical Denaturation

- **High salt concentration changes the ionic environment** of an enzyme, **disrupting ionic interactions** between different regions of the chain, while

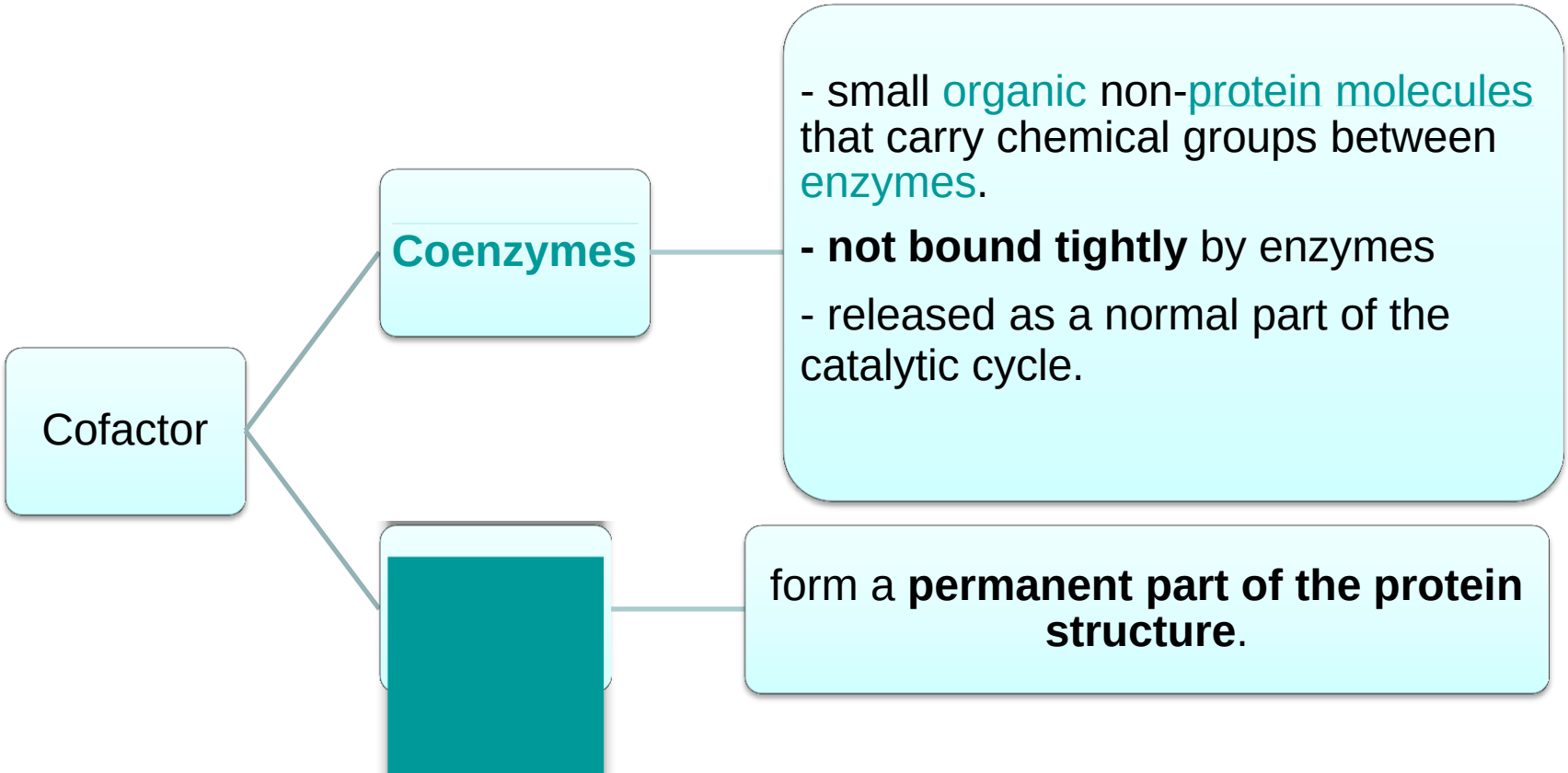
- **Urea denatures proteins by disrupting the hydrogen bonds** that maintain the secondary and tertiary structure of proteins.

- Certain chemical inhibitors totally inactivate enzymes; their effects are irreversible.

Cofactor

- A **cofactor** is a **non-protein chemical compound that is bound** (either tightly or loosely) **to an enzyme and is required for catalysis**.

- They can be considered "helper molecules/ions" that assist in biochemical transformations.



Prosthetic Groups

APOENZYME (protein molecule)	+	COFACTOR	→	HOLOENZYME (functional enzyme)

- For example, carbonic anhydrase, one of a group of metalloenzymes, contains a **metal ion at the heart of the active site**.

- In **carbonic anhydrase** the metal ion is a **Zn^{2+} ion**, and its presence is absolutely essential for enzymic activity.

Prosthetic Groups

- **Other enzymes have the haem group** as a cofactor. These include cytochrome oxidase, an enzyme involved in the crucial process of respiration.

- Cofactors such as these, which are an integral part of the enzyme structure.

Coenzyme

- Are **not integral parts of the enzyme structure**.
- They are **complex organic molecules**, often **derived from a vitamin**.
- These coenzymes, such as NAD^+ (nicotinamide-adenine dinucleotide), work together with the enzyme to bring about the required reaction.
- They **bind temporarily to the active site** of the enzyme and effectively **function as a co-substrate**, providing for the **transfer** of **groups** or **electrons** not readily available from the side-chains of the enzyme protein (Figure 1.29).
- They are released from the enzyme at the end of the catalysed reaction.

Coenzyme

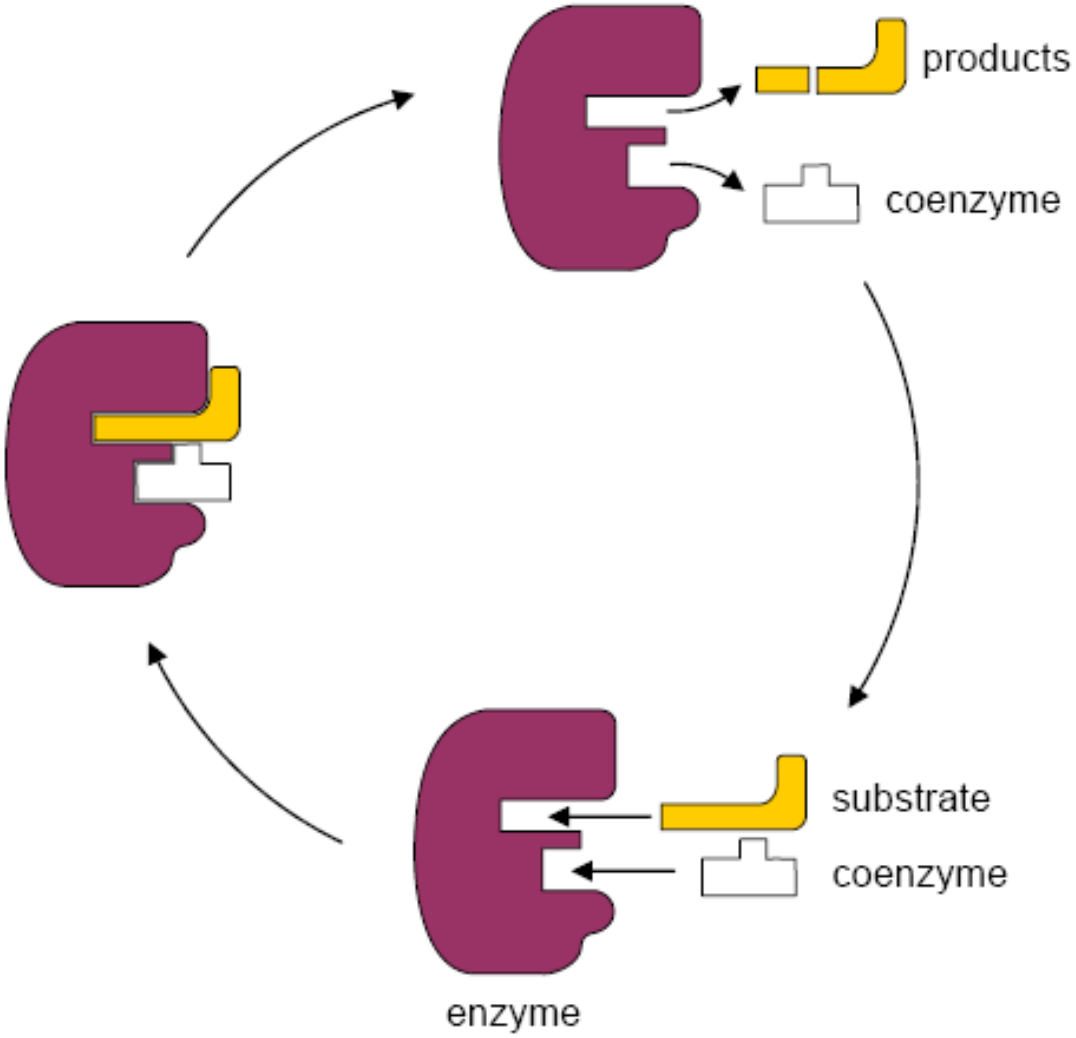


Figure 1.29 – binding of substrate and coenzyme to active site – the catalytic cycle

Coenzyme

- **NAD^+, $NADP^+$, and FAD** are important coenzymes because of their **ability to accept H^+ ions and electrons and therefore take part in redox reactions**.

- They are sometimes **referred to as 'hydrogen carriers'** because of their ability to transfer hydrogen atoms between reactions.

- Coenzyme A acts as a carrier of CH_3COO- groups and is therefore important in the metabolism of fatty acids

Coenzyme

- Both NAD^+ and FAD are derived from the water-soluble B group vitamins.

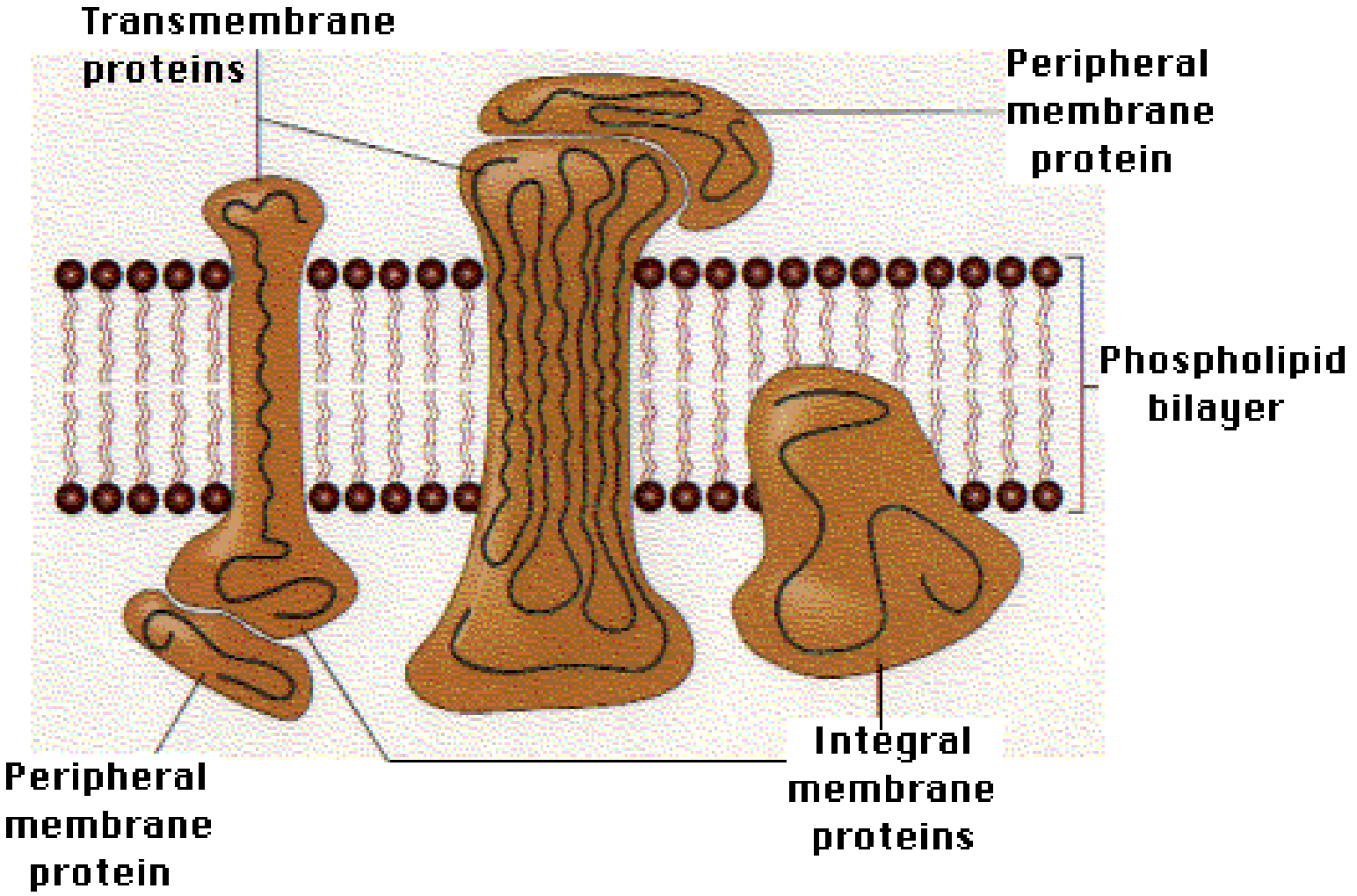
- In humans, an inadequate supply of these precursors gives rise to deficiency diseases.

Ion channels

- These channels are found to consist of protein subunits which sit across the plasma membrane – they are trans-membrane proteins.

- They **have enzyme-like function** and, by their **interaction with ions such as Na^+ and K^+** and relatively small molecules such as **ATP** (adenosine triphosphate), these proteins are able to selectively **control the transport of ions into, and out of, the cell**.

- In a manner similar to enzyme inhibitors certain plant alkaloids, oubain, for instance, can **interfere with ion transport** by **interacting with the protein units of the ion channels**.


Transmembrane proteins
Peripheral membrane protein
Phospholipid bilayer
Integral membrane proteins
Peripheral membrane protein