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that you need

WHEN IT COMES TO

Enzyme activity...

The good...

"CLINT COFACTOR"

...The Bad

"SEÑOR INHIBITOR"

"TERENCE TEMPERATURE"

AND THE "USEFUL"

Enzyme activity...

The good...

"CLINT COFACTOR"

Enzyme cofactors

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- Some enzymes need the presence of another molecule called a cofactor if they are to work effectively
- These are non-protein molecules that attach to the enzyme (**can be coenzymes or metal ions**)

They work in one of two ways:

- *They can modify the shape of the enzyme to make the optimum shape for substrate attachment*
- *They can participate in the reaction e.g. by attaching to a product for transfer to another enzyme*

Cofactors include organic molecules called **coenzymes:**

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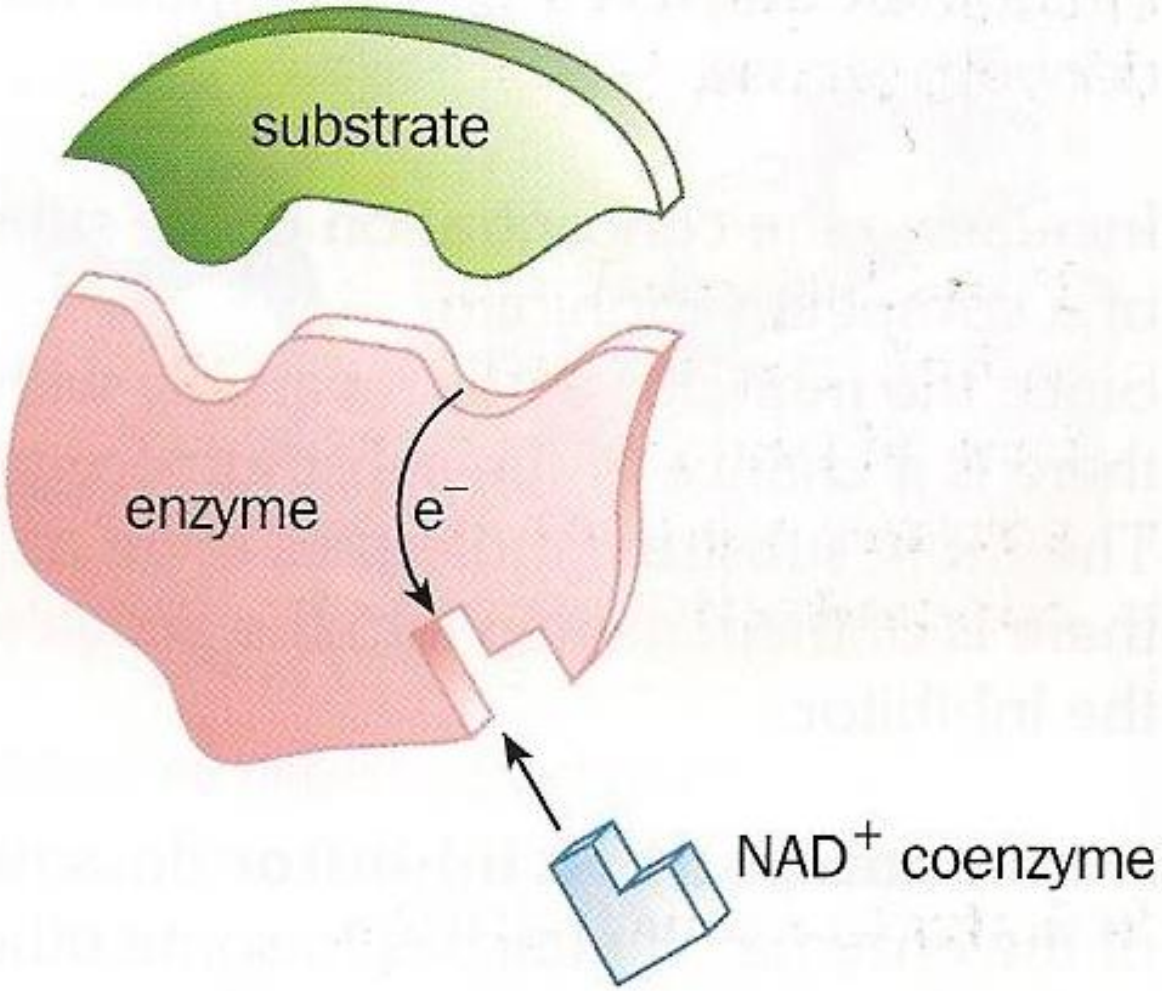
- These are small non-protein organic molecules that are not permanently attached to an enzyme. They help enzyme and substrate to bond with each other. The enzyme only functions if the coenzyme is present
- Many coenzymes are derived from vitamins e.g. NAD, *(from vitamin nicotinic acid, acts as a coenzyme for a number of dehydrogenase enzymes; it acts as a hydrogen acceptor. Involved in respiration reactions)*



NAD^+ is a coenzyme that works by removing an electron from the active site. The substrate is then able to engage

Cofactors also include **inorganic metal ions** (sometimes known as "activators")

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e.g. Mg^{2+}, Fe^{2+}, Ca^{2+} and Zn^{2+}. They form a temporary attachment to the enzyme active site to make a reaction more likely e.g. magnesium is needed as a cofactor for all protein synthesis in the body

Coenzymes and metal ions are mostly obtained from the vitamins and minerals in our diet

Fruit and vegetables are a rich source of vitamins and minerals

Table 4 Examples of cofactors and coenzymes

Enzyme	Cofactor	Role of enzyme
Carbonic anhydrase	Zinc ion (Zn^{2+})	Catalyses the combination of CO_2 with water to form carbonic acid in red blood cells, facilitating the transport of CO_2 in the blood
Cytochrome oxidase — a respiratory enzyme	Copper ion (Cu^{2+})	Combines electrons and hydrogen ions with oxygen in respiration
Enzyme	**Coenzyme**	**Role of enzyme and coenzyme**
Pyruvate decarboxylase— a respiratory enzyme	Coenzyme A	Pyruvate (3-carbon molecule) is broken down to acetate, which is 'picked up' by coenzyme A (forming acetyl CoA), and CO_2, which diffuses out of the cell
Succinate dehydrogenase — a respiratory enzyme	FAD (derived from vitamin B_2, riboflavin)	Hydrogen is removed from succinate and 'picked up' by FAD (to form $FADH_2$)

Tip The examples shown in Table 4 are provided only to illustrate the roles of cofactors and coenzymes. You do not need to learn these — most are involved in respiratory metabolism, which is covered in A2 Unit 2.

Enzyme activity...

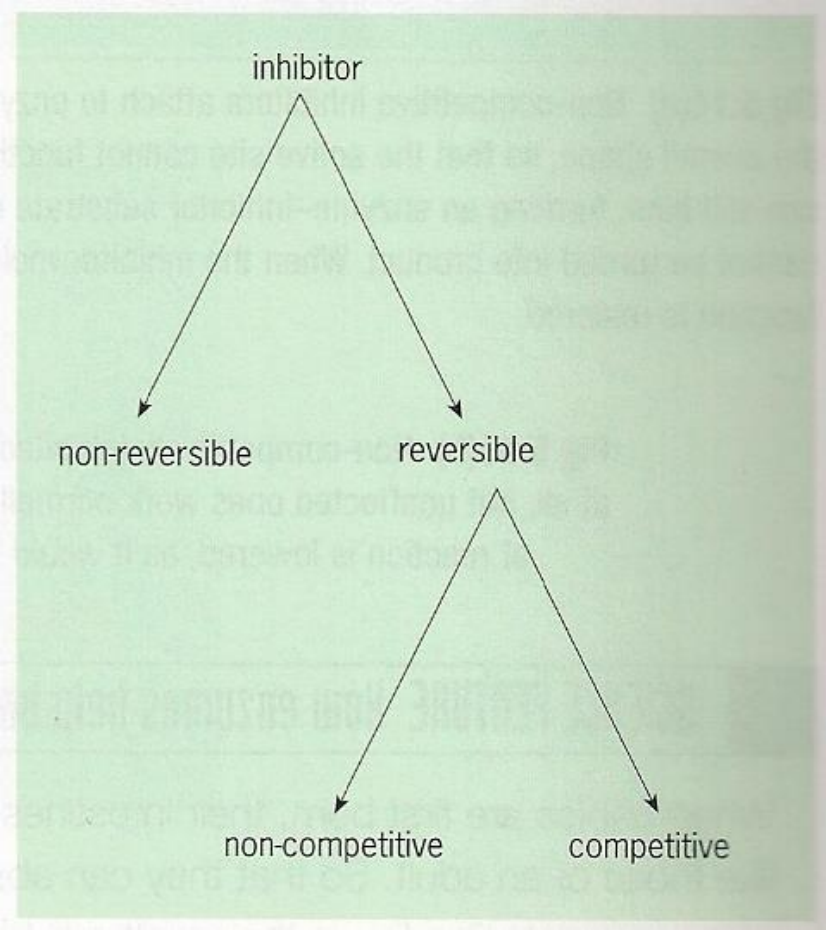
...The Bad

"SEÑOR INHIBITOR"

Enzyme inhibitors -

Inhibitors slowdown or stop enzyme reactions (they are reversible or non-reversible)

There are 2 types of **reversible inhibitors**: Competitive and non-competitive



- **Competitive:** Inhibitor has a similar structure to the substrate and competes to bind to the active site (doesn't remain there permanently)
- E.g. malonate is a competitive inhibitor of a respiratory enzyme succinate dehydrogenase

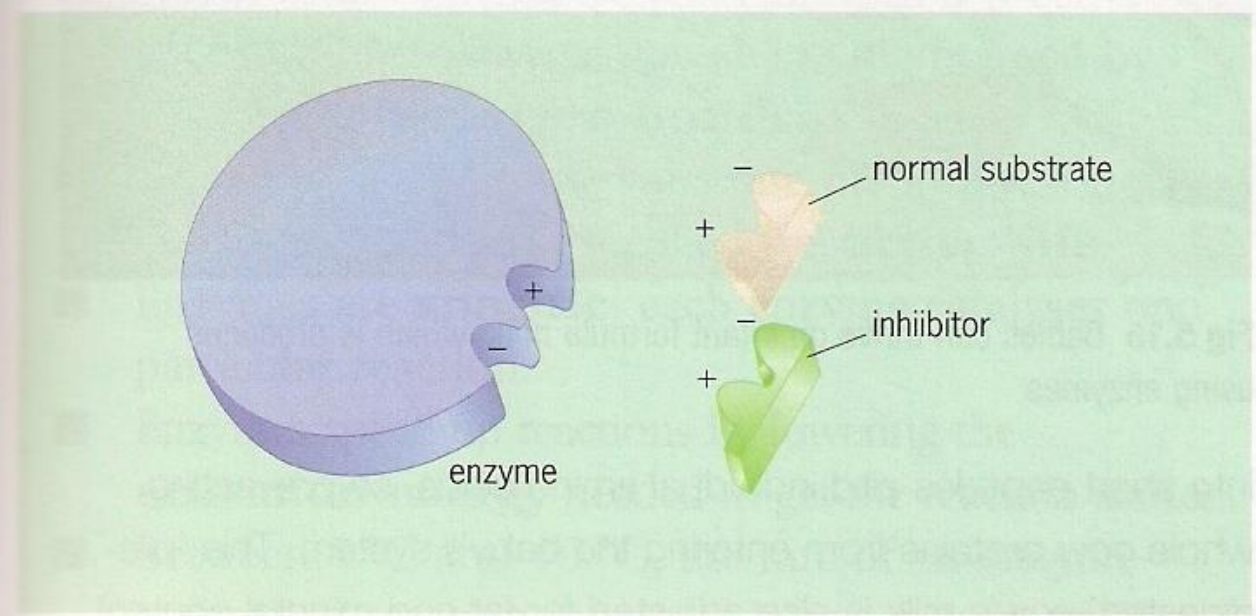
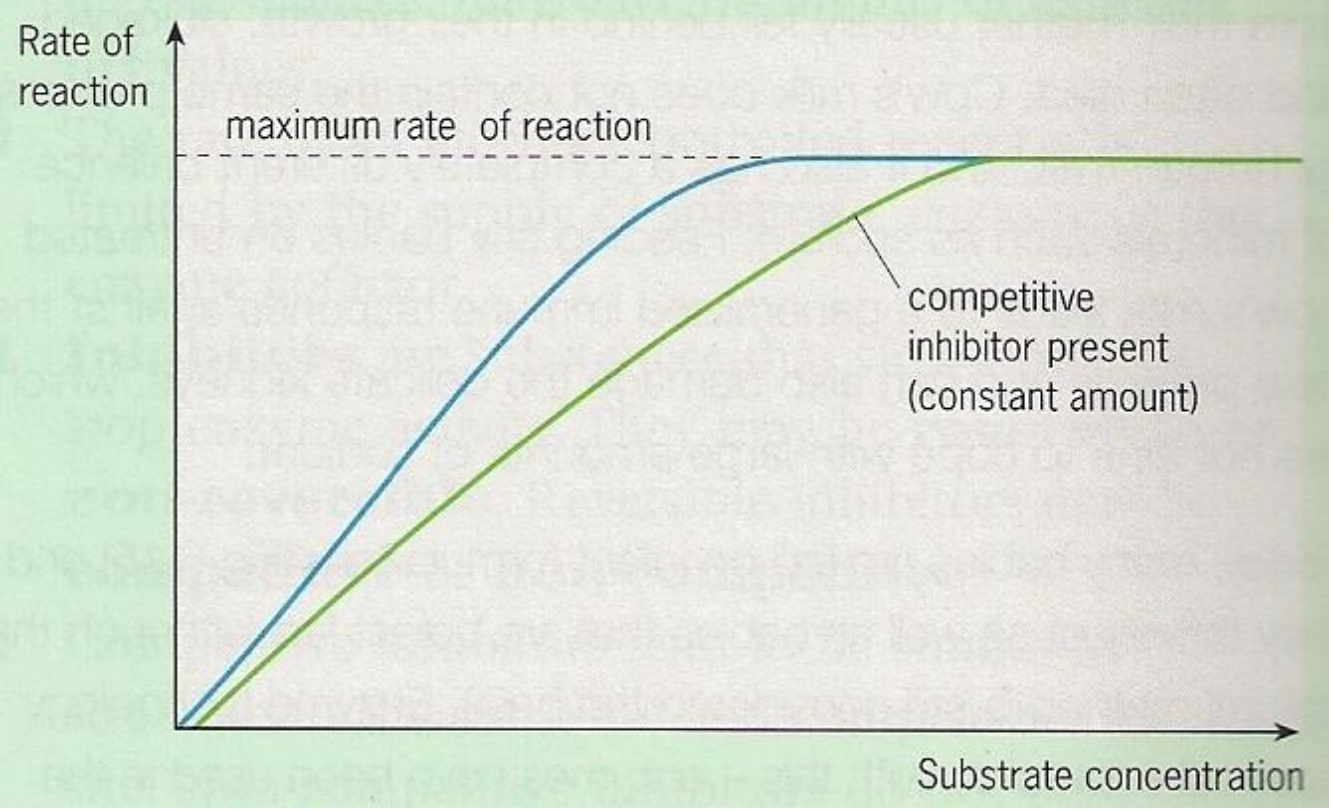


Fig 5.13(a) A competitive inhibitor fits into the active site of the enzyme, preventing the real substrate from gaining access. The inhibitor cannot be converted to the products of the reaction and so the overall rate of reaction is slowed down. If an inhibitor is present in equal concentrations to the substrate, and if both types of molecule bind to the active site equally well, the enzyme can only work at half its normal rate



- Increasing the substrate conc. reduces the effect of the inhibitor, as there is more chance for an enzyme substrate complex to form, rather than an enzyme-inhibitor complex
- Thus the degree of inhibition is related to the concentrations of inhibitor and substrate

Non-competitive: Binds to the enzyme, but not necessarily at the active site to change the shape of the active site or block it

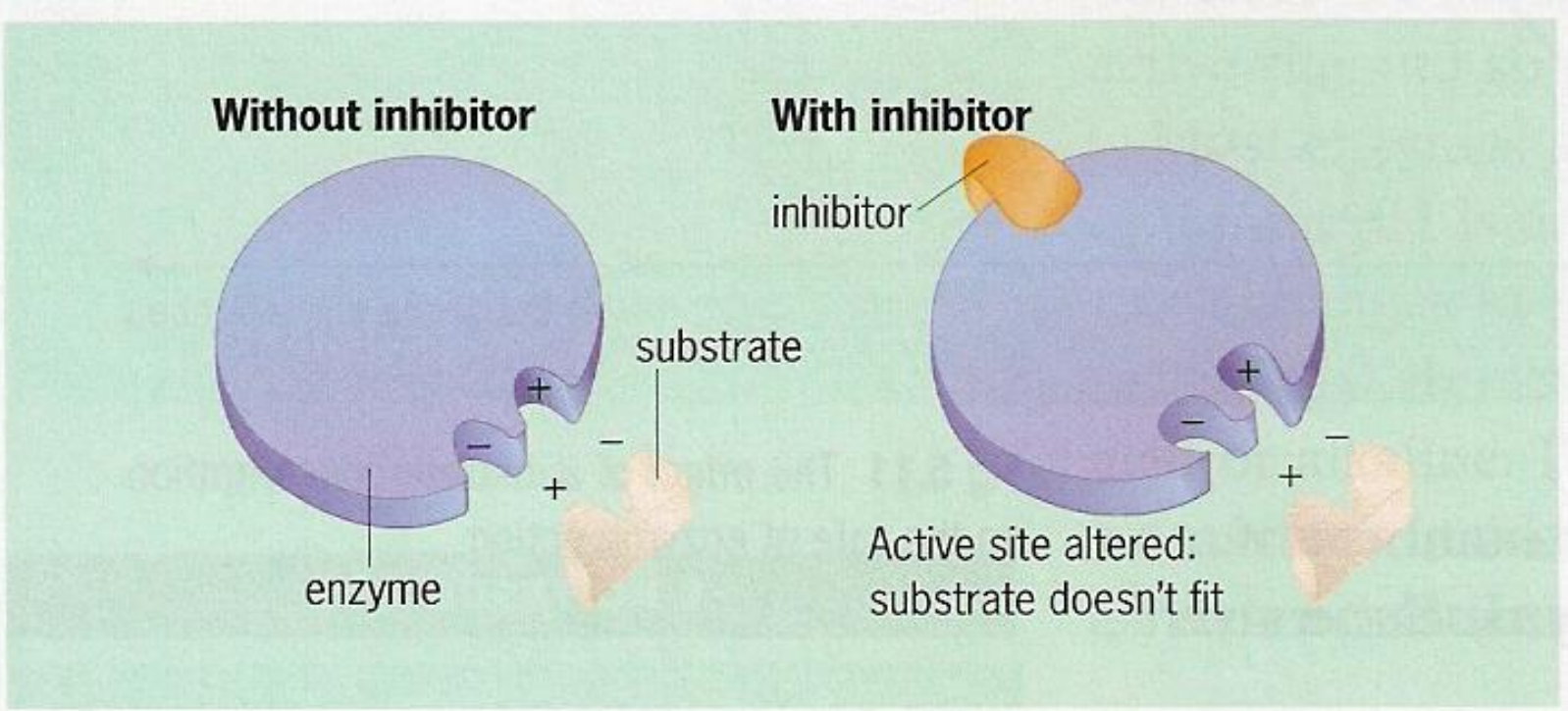
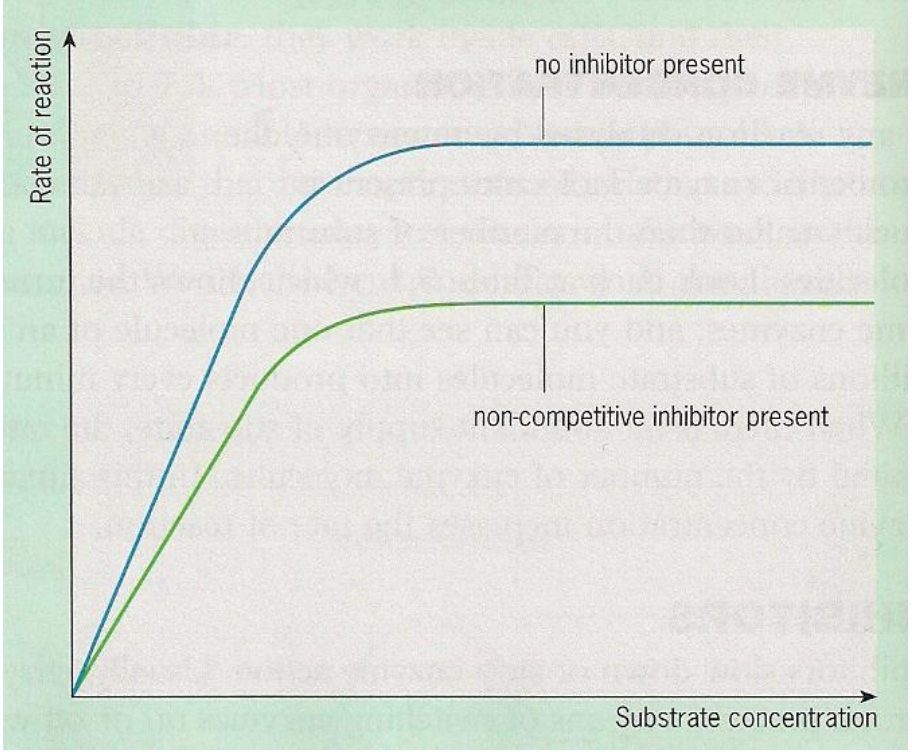


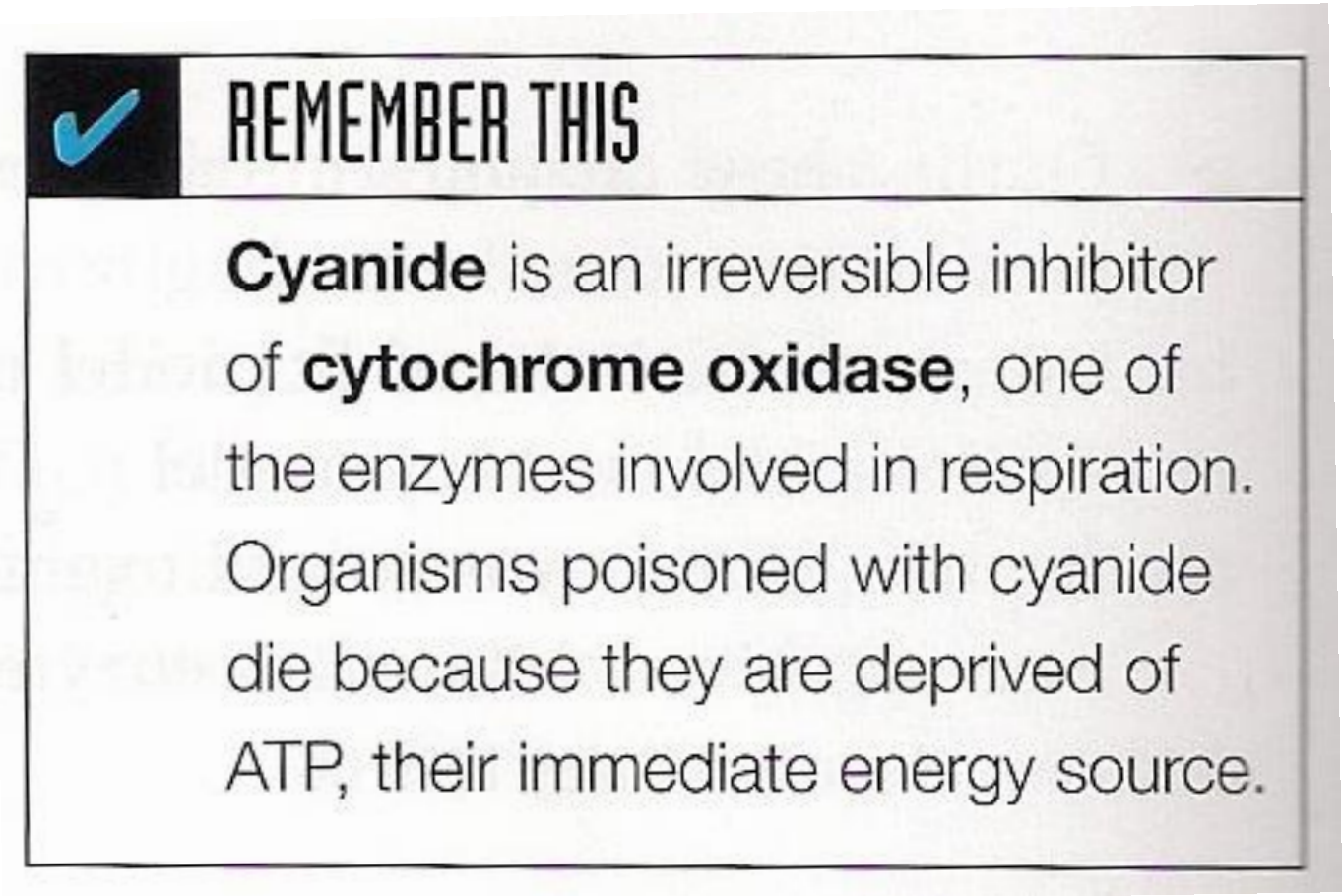
Fig 5.14(a) Non-competitive inhibitors attach to enzyme molecules and alter the overall shape, so that the active site cannot function. Although the substrate can still bind, forming an enzyme–inhibitor substrate complex, the substrate cannot be turned into product. When the inhibitor molecule is removed, normal function is restored



- This can sometimes be irreversible (see cyanide e.g.)
- The enzyme is allosteric i.e. the enzyme has two shapes, an active one and an inactive one depending on whether the inhibitor is bound
- Adding more substrate won't reduce a non-competitive inhibitors effect as there is no competition for the active site

Non-reversible inhibitors change the tertiary structure of the enzyme by binding to it permanently.

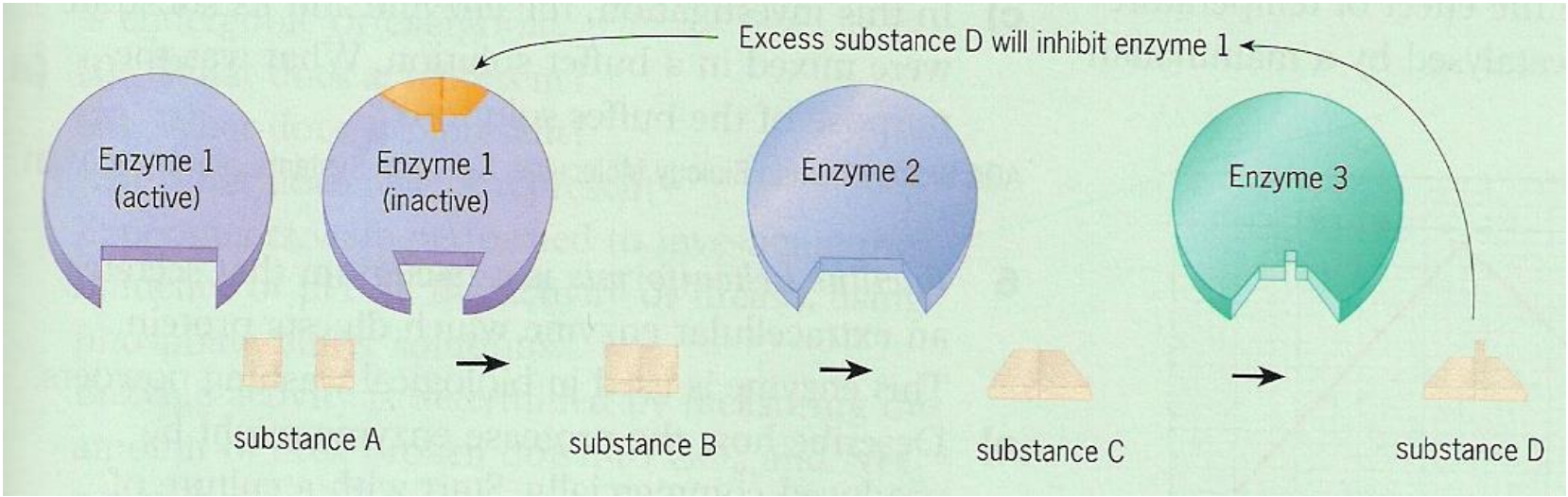
- This destroys its substrate specific shape e.g. cyanide, a non-competitive irreversible inhibitor:



REMEMBER THIS

Cyanide is an irreversible inhibitor of **cytochrome oxidase**, one of the enzymes involved in respiration. Organisms poisoned with cyanide die because they are deprived of ATP, their immediate energy source.

Negative feedback:



Metabolic pathways consist of a series of enzyme controlled reactions i.e. the product of the reaction is the substrate for the next reaction

Often an end product is a non-competitive inhibitor for an enzyme earlier in the chain (so as to prevent too much being made)

This is negative feedback (the output affects the input)

Enzyme activity...

"TERENCE TEMPERATURE"

THE "USEFUL"

TELL ME WHY...

Temperature can be a useful factor for enzyme action.

By not being free in solution there are many advantages but also some disadvantages...

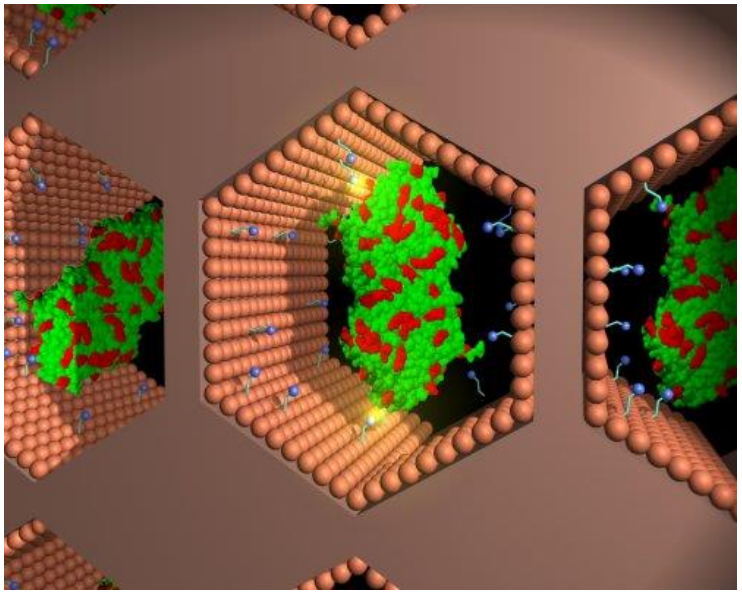
Advantages:

- Avoids contamination of the product with enzyme
- Immobilisation can make enzymes more stable/tolerant of a greater range of pH and temperature (thermostability) than it would be if it were just in solution
- The enzyme can be reused and is easier to recover at the end of the process (less expensive than enzyme solutions)
- Allows for continuous use

Disadvantages:

- The active site may be blocked by the support matrix (e.g. in adsorption)
- Insoluble substances might slow the substrate from reaching the enzyme (e.g. in entrapment)
- The active site may be altered during the binding process (e.g. in cross linkage)
- Immobilisation process can be expensive and complicated

Entrapment: →



The flow rate through the column also influences the activity of the enzyme:

-If it is too slow the reaction is completed too early in the column

-If it is too fast then not all of the substrate will react with the enzyme

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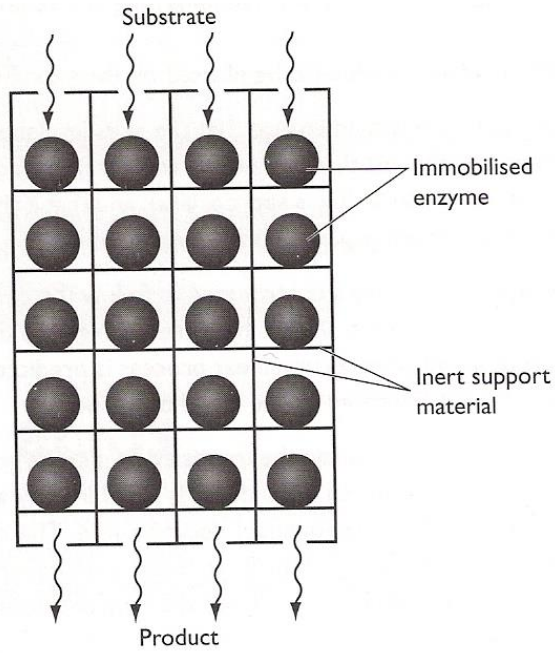

Figure 23 Immobilised enzymes in a continuous-flow column reactor

Industrial /commercial uses of immobilised enzymes:

- Immobilised onto reagent strips for diagnostic (diagnosis) purposes e.g. glucose oxidase and peroxidase on clinstix
- Using immobilised lactase to produce lactose free milk for cats (most cats are lactose intolerant)
- Turning inexpensive corn starch into high fructose corn syrup (very sweet, used in soft drinks and confectionery)
- Washing powders contain immobilised proteases

Read the immobilised enzymes practical