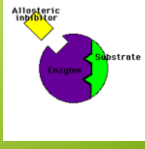
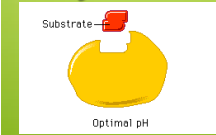
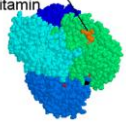


Molecular Biology

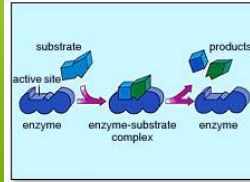
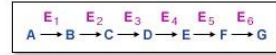
2.5- Enzymes

Co-Enzyme or Vitamin

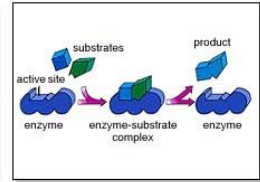


Essential idea:

- Enzymes control the metabolism of the cell.



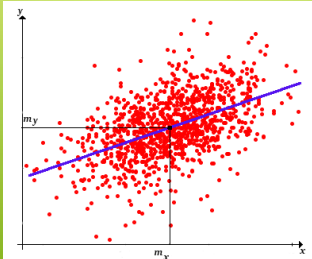
Degradation Reaction



Synthetic Reaction

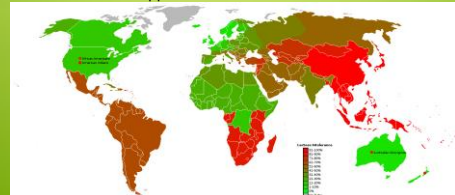
Nature of science:

- Experimental design
 - accurate, quantitative measurements in enzyme experiments require replicates to ensure reliability. (3.2)



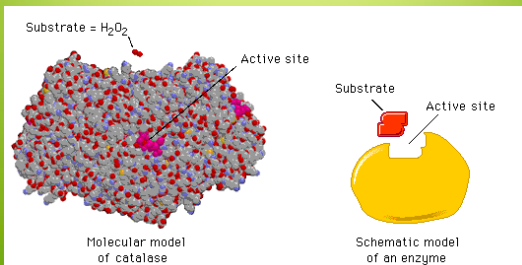
Theory of knowledge:

- Development of some techniques benefits particular human populations more than others.
 - Example: the development of lactose-free milk available in Europe and North America would have greater benefit in Africa/Asia where lactose intolerance is more prevalent. The development of techniques requires financial investment.
 - Should knowledge be shared when techniques developed in one part of the world are more applicable in another?



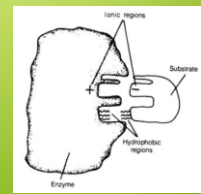
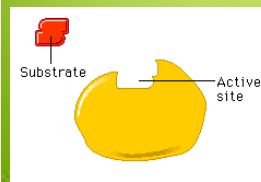
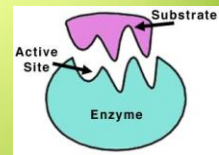
Enzymes

- Enzymes are globular proteins
- Enzymes have an active site to which specific substrates bind.



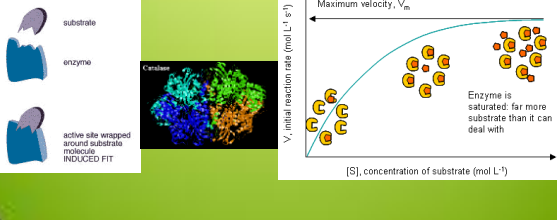
Enzyme-Substrate Complexes

- Substrates are reactants in an enzymatic reaction.
- The substrate must be complementary to the surface properties (shape and charge) of the active site.
- Active site is a microenvironment



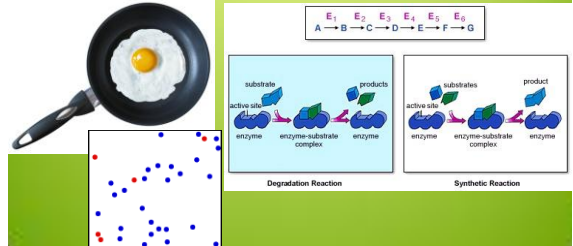
Enzyme-Substrate Complexes

- Induced-fit model
 - Slight change in enzyme shape when substrate binds
 - Facilitates the reaction
- When all enzymes are filled (saturated) reaction can't go faster
- Most enzymes named adding the ending "-ase."



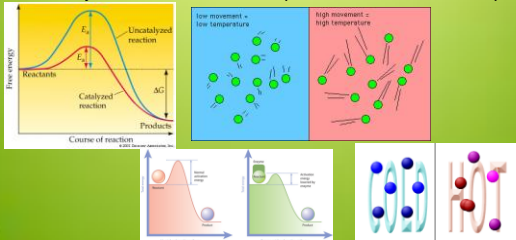
How Enzymes Work

- Enzymes are **catalytic** proteins
- Speed chemical reactions without being changed
- Enzyme catalysis involves molecular motion and the collision of substrates with the active site.
- Every enzyme catalyzes only one reaction or one type of reaction.



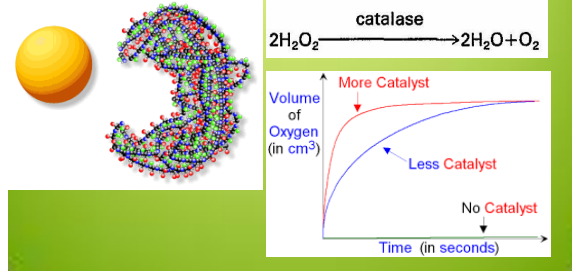
How Enzymes Work

- Enzymes lower the Energy of Activation
 - Energy of activation (E_a)** is energy that must be added to cause molecules to react
 - Heat speeds a reaction, but denatures proteins
 - Enzymes allow reactions to proceed at moderate temps



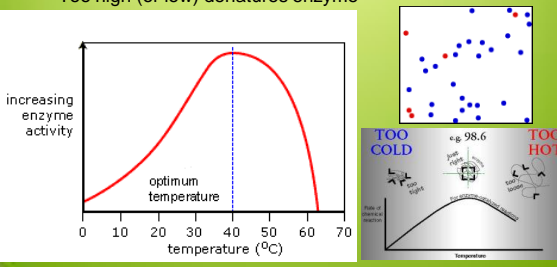
Enzyme Activity

- Enzymatic reactions are rapid
 - Most occur 1000 times/sec
 - 2H₂O₂ → 2H₂O + O₂ (600,000 times/sec with catalase).



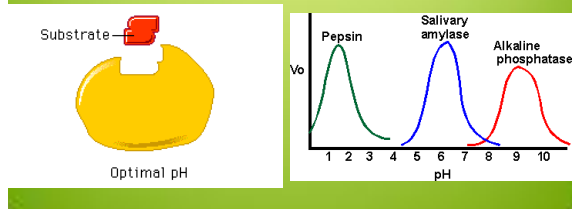
Enzyme Activity

- Temperature
 - Increase temp → increase molecular collisions → increase enzyme activity
 - Too high (or low) denatures enzyme



Enzyme Activity

- pH
 - Each enzyme has optimal pH that maintains its normal configuration.
 - A change in pH alters ionization of side chains, eventually resulting in denaturation.
 - Optimal pH varies per enzyme



Enzyme Activity

- Concentration
 - Enzyme- increases rate (to a point)
 - Substrate- increases rate till enzyme is saturated

Rate of reaction

Enzyme concentration

Maximum velocity, V_m

V_0 initial reaction rate ($\text{mol L}^{-1} \text{s}^{-1}$)

[S] concentration of substrate (mol L^{-1})

Enzyme is saturated: far more substrate than it can deal with

Cofactors and Coenzymes

- Many enzymes require an inorganic ion or nonprotein cofactor to function
- The enzyme may only become active when all the appropriate cofactors or coenzymes are present and bind to the appropriate sites on the enzyme.
- Cofactors- inorganic ions (iron, zinc, copper)
- Coenzymes**- Organic cofactors (vitamins)

Controlling Metabolism

- Other molecules and the environment in which the enzyme acts can enhance or inhibit enzyme activity.
- Molecules can bind reversibly or irreversibly to the active or allosteric sites, changing the activity of the enzyme.

TOO COLD e.g. 98.6 TOO HOT

Rate of reaction

Temperature

(a) Reaction

Substrate Enzyme Enzyme binds substrate Enzyme releases products

(b) Inhibition

Inhibitor Enzyme Enzyme binds inhibitor Inhibitor competes with substrate

Controlling Metabolism

- Competitive Inhibition
 - Another molecule is similar to enzyme's substrate
 - Competes** with substrate for enzyme's **active site**
 - Decreases product formation rate.

No inhibitor present - the substrate binds

Competitive Inhibition

$E + S \rightleftharpoons ES \rightarrow E + P$

$E + I \rightleftharpoons EI$

Rate of reaction

Substrate concentration

Without inhibitor

With competitive inhibitor

Controlling Metabolism

- Allosteric Interactions
 - Noncompetitive Inhibition
 - A molecule binds to an allosteric site (a site other than active site)
 - Changes the three-dimensional structure of the enzyme
 - Cannot bind to its substrate.
 - Allosteric Activation

Allosteric inhibitor

Enzyme

Substrate

Active site (correct conformation for substrate)

Substrate

Active site (correct conformation for substrate)

Inhibitor site

Allosteric inhibitor

Allosteric activator

Activator site

Schematic representation of allosteric enzyme activity

Substrate

Active Site

Enzyme

Regulatory Site

Allosteric Activator

Controlling Metabolism

- Feedback Inhibition
 - Regulates activity of most enzymes
 - Product binds to enzyme's active or allosteric site
 - Concentrations of products can be kept within narrow ranges.

substrate

active enzyme

end product

metabolic pathway

inactive enzyme

end product binds to enzyme

Negative feedback

Enzyme 1

Enzyme 2

Enzyme 3

Product A

Product B

(a) Negative feedback

Σ first enzyme in pathway

Denaturation of Enzymes

- Denaturation of proteins by heat or by deviation of pH from the optimum can immobilize proteins
 - Remember these slides? (from 2.4- Proteins)

Immobilized Enzymes

- Enzymes used in industry are usually immobilized.
- They are attached to a material so that their movement is restricted.
- Common ways of doing this are:
 - Aggregations of enzymes bonded together
 - Attached to surfaces, e.g. glass
 - Entrapped in gels, e.g. alginate gel beads

Immobilized Enzymes

- Widely used in industry.
 - Detergents contain proteases and lipases to breakdown protein and fat stains.
 - Breakdown the starch or cellulose into biofuels that can be combusted.
 - In the textiles industry enzymes help in the processing of fibers and leather.
 - Paper production uses enzymes to helping in the pulping of wood

Immobilized Enzymes

- Widely used in the food industry
 - Pectin to increase the juice yield from fruit.
 - Rennin is used to help in cheese production.
 - Various enzymes used in clarification of beer.

Immobilized Enzymes

- Medicine & Biotechnology
 - Diagnostic tests
 - Contact lens cleaners
 - Restriction enzymes to cut DNA in genetic engineering.

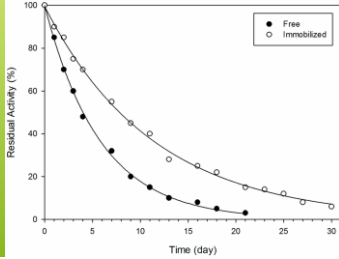
Serum Enzymes	Location of serum enzymes	Concentration increased in	Concentration decreased in
Lipase	Pancreas	Acute pancreatitis, Pancreatic carcinoma	Liver disease, vit-A deficiency, diabetes mellitus
Amylase	Saliva	High intestinal obstruction, Acute pancreatitis, Parotitis, Diabetes	Liver disease
Trypsin	Stomach	Acute disease of pancreas	-
Cholinesterase	-	Nephrotic syndrome	Liver disease, Malnutrition
Alkaline phosphatase	Bone, liver	Rickets, Jaundice, Metastatic carcinoma, Kidney disease	-
Acid phosphatase	Prostate	Metastatic prostatic carcinoma	-

Advantages of Enzyme Immobilization

- Concentration of substrate can be increased as the enzyme is not dissolved
 - this increases the rate of reaction
- Recycled enzymes can be used many times
 - Easy to separate from the reaction mixture
 - Reaction can be stopped at the correct time
 - Cost saving.

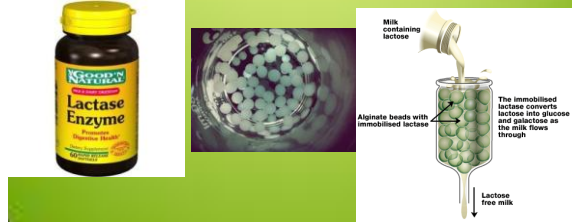
Advantages of Enzyme Immobilization

- Stability of the enzyme to changes in temperature and pH is increased
 - Reduces the rate of degradation
 - Cost saving.



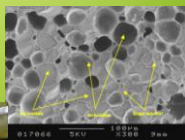
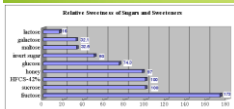
Production of Lactose-free Milk

- Lactase obtained from commonly from yeast or bacteria.
- Lactase is bound to the surface of alginate beads.
- Milk is passed (repeatedly) over the beads.
- The lactose is broken down into glucose and galactose.
- The immobilized enzyme remains to be used again



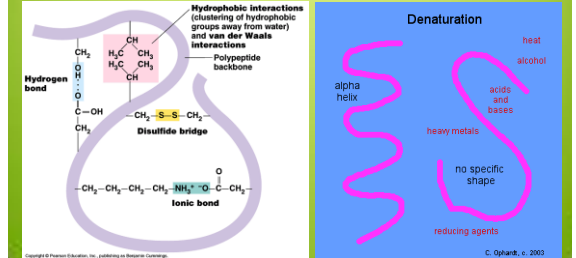
Production of Lactose-free Milk

- Other uses of lactose free milk:
 - Increase the sweetness of milk (glucose and galactose are sweeter)
 - Reduce need for artificial sweeteners
 - Reducing the crystallization of ice-creams (glucose and galactose are more soluble than lactose)
 - Shorten the production time for yogurts or cheese (bacteria ferment glucose and galactose more readily than lactose)



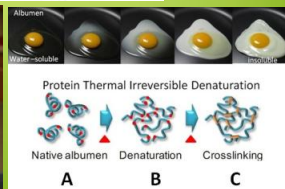
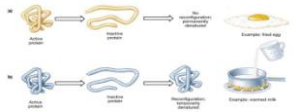
Altering Protein Structure

- Most bonds and interactions of protein structure are relatively weak can be disrupted or broken.
- This results in a change to the conformation of the protein, which is called denaturation.



Altering Protein Structure

- Denaturation is permanent in most cases.
- Soluble proteins often become insoluble and form a precipitate.



Altering Protein Structure

- Extremes of pH can cause denaturation
 - charges on R groups are changed
 - breaks ionic bonds within the protein or causes new ionic bonds to form.
- Heat can cause denaturation
 - vibrations within the molecule breaks intermolecular bonds or interactions.

