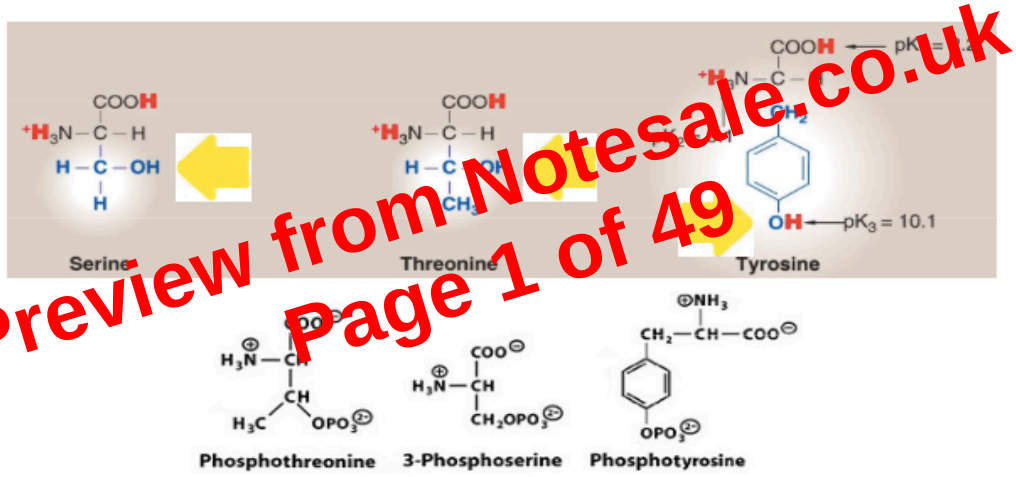
Biochemistry lecture notes

**Lecture 1:**

- Central dogma: cell information flows from DNA→ RNA→ Protein
- DNA replicates, then gets transcribed into RNA, and then translated into proteins (chain of amino acids)
- 20 major amino acids
    - only L forms are found in our cells, D-forms are found in bacterial cell walls
    - 4 classes of amino acids:
        - Polar: negative charge/ acidic
        - Polar: positive charge/ basic
        - Polar: uncharged
        - Non-polar
    - The properties of the side chain that determine the protein's character include:
        1. Hydrophobic (non-polar) or hydrophilic (polar)
        2. Physical size
        3. Ability to form hydrogen bonds






Phosphorylation of these 3 amino acids can have a dramatic effect on protein function. How to test in the laboratory? We assume phosphorylation is essential to function.

1. Replace amino acid thought to be phosphorylated with alanine (charge = 0). What happens to the function of the protein?
2. Replace amino acid with an acidic amino acid, like aspartic acid (charge = -1). What happens to the function of the protein?

*Acidic amino acids mimic phosphorylation.*

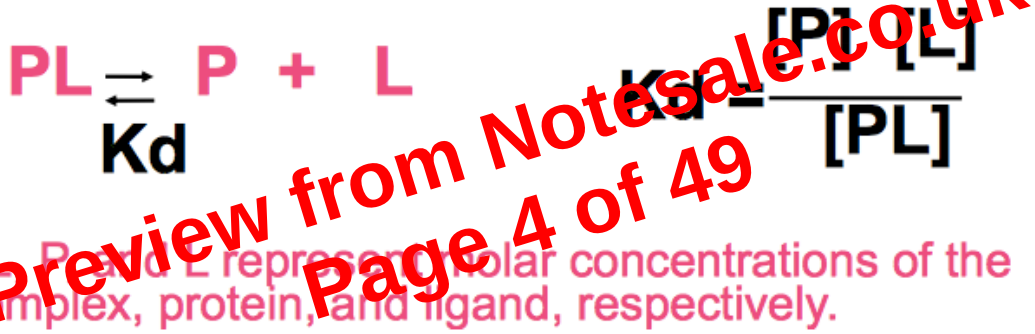
*NEED TO KNOW PROPERTIES, 3 LETTER AND 1 LETTER CODES, AND STRUCTURE OF 20 AMINO ACIDS

**Lecture 2:**

- Protein structure:
    - **Primary structure:** peptide bond (covalent dehydration, between carboxyl group and amino group)
    - **Secondary structure:** Hydrogen bonding (beta or alpha sheets)

- Amino acid residues that are necessary for function are found in all organisms→ thus, a common protein sequence in a all organisms means that it's a protein from evolution that has been conserved because it is necessary for organism's function
  - Called "conserved residues"
  - Example: steroid hormone receptor

- **Ligand binding proteins:**
  - Many proteins contain sites to which ligands specifically bind and form a complex with the protein
  - Ligand: molecule that can form this complex
  - Binding occurs by multiple weak or a few strong forces
  - Ligand binding is extremely specific to the protein's binding site
  - Example: hemoglobin is an iron-containing oxygen transport metalloprotein in the red blood cells of all vertebrates—it binds specifically to iron in order to carry its function of transporting oxygen
- **Ligand binding-dissociation constant**
  - **Dissociation constant (Kd):** a measure of the affinity of an interaction between a ligand and a protein
  - An equilibrium constant
  - For a protein (P) binding ligand (L)






  - High affinity=tight binding= small Kd value
  - Units are M (concentration)

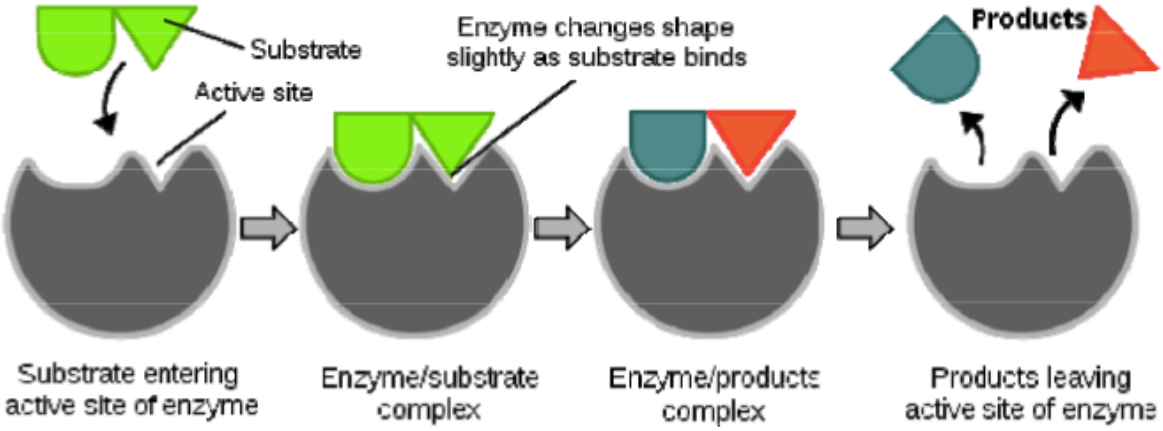
****Remember the smaller the Kd value, the higher the affinity!!**

- The ribosomal complex is made up of RNA, protein, and tRNAs

**Lecture 5: Collagen and Antibodies**

**Collagen:** 300nm long & 1.5nm diameter protein

- Most common protein in animals
- Extracellular, forms strong fibers or sheets
- Connects and strengthens tissues
  - Found in bone matrix, tendons, connective tissue, skin
- Characteristic structure triple helical
- Found in marketplace in face creams, plastic surgery injections (to fix wrinkles, get fuller lips etc.), collagen drinks, acne treatments
- **Collagen helical folding:**
  - Presence of so much proline prevents alpha helix
  - Instead, forms a poly-proline type 2" helix
    - More extended than alpha helix
    - No intra-chain H bonds




- Kinetics vs. Equilibrium:
  - Enzymes DO NOT change equilibrium (the final concentration of substrate and product), nor do they change the free energy (G) of the reaction
  - Rather, enzymes only affect the speed/kinetics of the reaction, allowing equilibrium to be achieved sooner
  - Both forward and reverse reactions are catalyzed by an enzyme
  - Enzymes also do not change the free energy difference between substrate and product
- Mechanisms to lower activation energies:

1. Enzyme binds to two substrate molecules and **orients** them precisely to encourage a reaction to occur between them
2. Binding of substrate to enzyme rearranges electrons in the substrate, creating **partial negative and positive charges** that favour the reaction
3. Enzyme **strains** the bound substrate molecule, forcing it toward a transition state to favour a reaction



Transition state analogues:

- Compounds that resemble the transition state, but do not undergo a chemical reaction
  - Similar geometry, charge distribution etc.
- Good inhibitors because they bind tightly, blocking the active site→ **competitive**
- Inhibitors can also bind to other sites of the enzyme→ **non-competitive**
- Many drugs and antibiotics are enzyme inhibitors, and are most effective if they are transition state analogs
  - Example: Oseltamivir (Tamiflu) flu medication is a neuraminidase inhibitor—prevents flu virus from releasing itself from the host cell

Apoenzymes and cofactors:

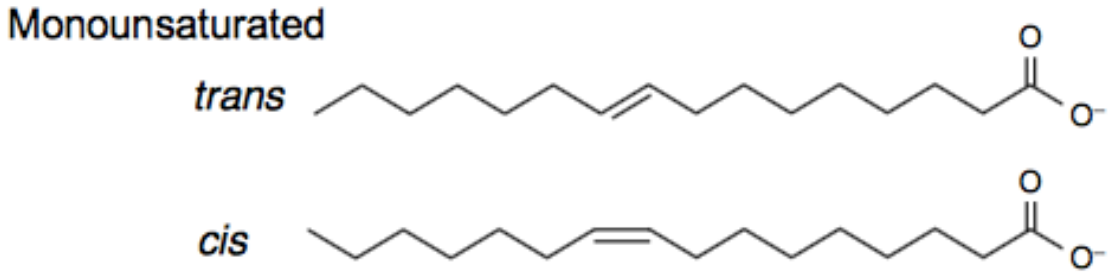
- **Apoenzymes:** inactive enzymes that require a partner (cofactor) to function, thus activate it to become a holoenzyme
- **Cofactor:** non-protein molecule that binds to apoenzyme in order to activate it
  - Co-factors can either be essential to enzyme function or simply stimulate its function causing the rate of reaction to increase even further
  - Co-factors can be organic or metal ions (iron, zinc)
- **Holoenzyme**: whole enzyme—apoenzyme+ cofactor, thus it is the enzyme is an active state

Enzyme optimal conditions:

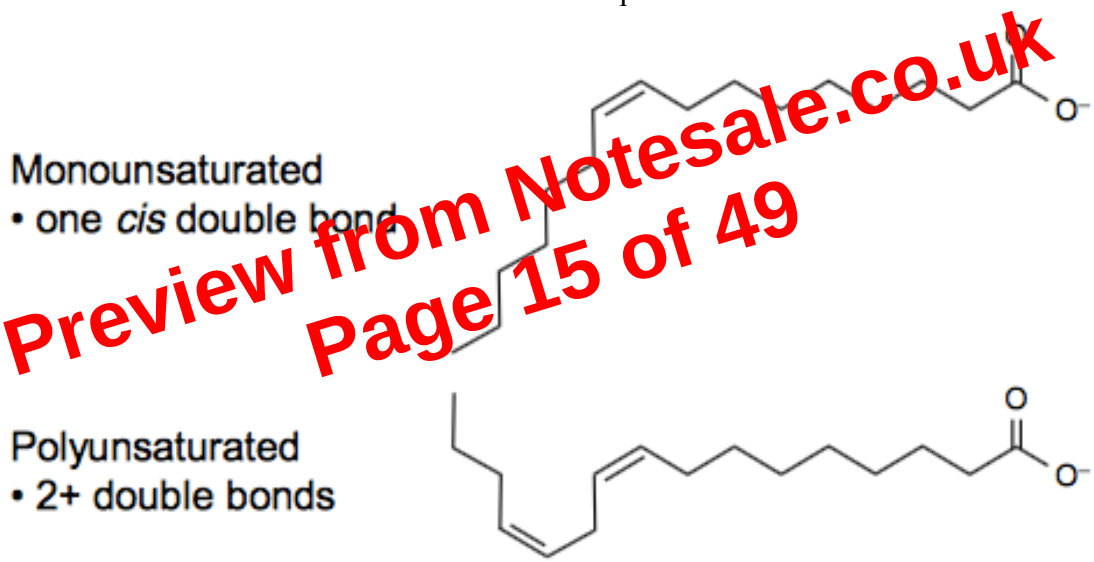
- Enzymes function best under optimal conditions called “optimal temperature or pH”

**TOPIC 9: Lipids and biological membranes**

- **Lipid:** a biological that is insoluble in water
- **Fatty acids:** used for energy and structure
  - A long hydrocarbon chain with a carboxylic acid at the end
  - **Saturated:** no double bonds, all carbons are single bonded to H and other C
  - **Unsaturated:** 1 or more double or triple bonds




- In living organisms, cis fatty acids are most dominant
- The first double bond normally goes between the 9th and 10th carbon (not random usually)
- Cis forces the chain to have a bend in it—real representation:






- Short forms to refer to fatty acids:
  - 16:0 is a linear saturated 16 carbon chain
  - For unsaturated (all these options can name the same thing):
    - 16: 1c Δ9 (1 cis bond at the 9th C)
    - 16:1 (n-7)
    - 16:1 (w-7)
    - 16:1
- fatty acid saturation and blood cholesterol
- LDL= low density, lipoprotein, carries cholesterol to tissues
- HDL= high density lipoprotein, scavenges cholesterol from tissues

| Dietary Fat | Effect on LDL | Effect on HDL | Overall effect |
|---|---|---|---|
| saturated | increase | increase | even |
| Cis-unsaturated | decrease | increase | good |
| Trans-unsaturated | increase | decrease | bad |

        - At this point G=0
    - **Le Chatelier's principle:**
        - If an external force acts on the system, reaction shifts to counteract changes in concentration in order to return back to equilibrium
        - Demonstrates that reaction is reversible-- **Sensitive to changes in product/reactant concentrations
        - For example, if add more products to the system, the system will shift to favour the reactant sides—thus making more reactants to level out their concentrations

**Irreversible reactions:**

- Spontaneous, energetically favourable
- Products are extremely strongly favoured—have an extremely lower G value (very negative) than reactants
- The backward reaction is so uncommon that in essence it only goes in one direction; reactants→ products
- Insensitive to changes in concentration
    - No matter what you do, it doesn't matter because the G value of the products continue to stay very negative
- If a reaction does not proceed to equilibrium, it indicates that it is an irreversible reaction

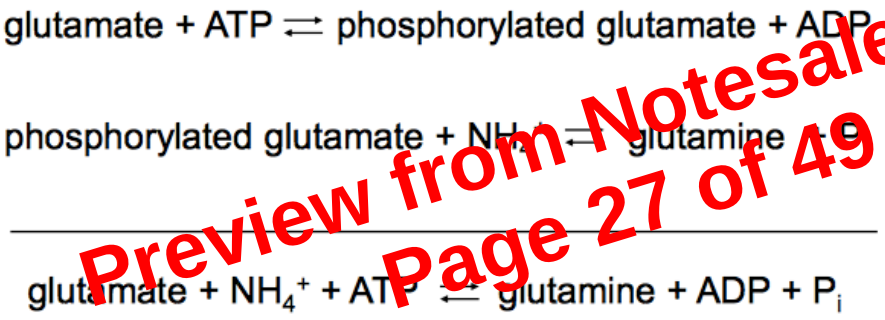
- **Energetically unfavourable reaction:**
    - Reactants are favoured, they have a lower G value than the products
    - Non-spontanous, need energy input in order to make it happen
    - Changing concentrations enough to favour products may not be possible
    - Enzymes change the kinematics of a reaction, however do not change the thermodynamics of the reaction—thus, the enzyme does not make a reaction spontaneous, it just helps it proceed!!
        - Enzymes help reaction faster but do not change the direction of reaction
        - They lower the activation energy but do not actually change the relative G
        - If a reaction is favourable in backwards reaction, it will simply go faster in the backward direction
        - Thus, enzymes can ONLY be used with a energetically favourable forward reaction if you desire the the forward reaction to happen
    - This is where reaction coupling solves the problem of making unfavourable reactions become favourable
    - An energetically favourable reaction can be used to make unfavourable reaction proceed
    - The reactions must be chemically coupled together
    - Favourable reaction is one with a negative G value, thus coupling a very negative reaction and a smaller positive reaction will result in a negative G value
    - However, you do not want to use way too much energy because most of the energy would be wasted as heat
    - Thus want to find a reaction that has a negative value close to the positive value of the reaction of interest
    - Cells store energy from food in carrier molecules, to be spent a little energy at a time—conserving their energy

3. Since molecule is very hydrophilic, it really likes to associate with water, you can fit more water molecules around the products than the reactants—the products are more highly solvated with water than the reactants
4. The ratio of ATP is much higher than ADP in the cell, thus, this makes making ADP from ATP much more favourable

**ATP is said to have "high-energy" phosphoanhydride bonds because of it's position relative to
- It's in a position that if you let it go, it will go from high energy to low energy spontaneously
- Bond is situated in the molecule in a way that makes is in a high energy state

→ Analogy: a marker itself does not have any more energy than other identical markers of the same make, colour etc. however, if you position it up high in the air and the rest are on the floor, it has high energy and it will want to fall down to the floor into its low energy state—thus, only high energy because of where it is situated

- ATP-driven formation of glutamine:
  - Know the mechanism
  - Coupling ATP hydrolysis with synthesis of glutamine
  - Take glutamic acid combined with ammonia to make glutamine
  - Has a high-energy intermediate
  - Cannot separate the hydrolysis of ATP from the addition of ammonia—this is what coupling is—their mechanisms are intertwined






$$\Delta G^\circ = +3.4 \text{ kcal/mol} - 7.3 \text{ kcal/mol}$$
$$= -3.9 \text{ kcal/mol}$$

**Still wasting some energy (-3.9kcal/mol) however this is minimal compared to the use of other molecules which is why ATP is often used

What if we need more energy than what ATP→ ADP supplies?
- Option 1: Hydrolysis ATP to AMP and 2 Pi
  - Gives you a little more than double the energy (-15.8)
- Option 2: Couple to a reaction that releases more energy than ATP
  - Example: phosphocreatine has a -10.3kcal/mol, results in creative and inorganic phosphate

Note of Kinetics vs. thermodynamics:
- Remember that knowing the G value does not tell you the kinetics (speed) of the reaction

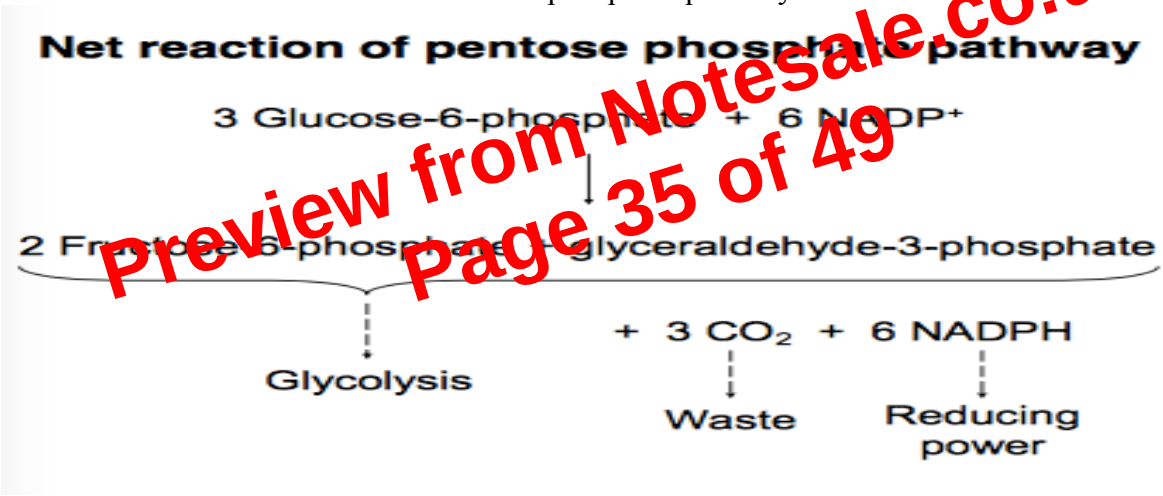
Reduction-Oxidation Reactions:
- Transfer of electrons from one to another, need a pair
- Some biochemical reactions require a molecule to be reduced

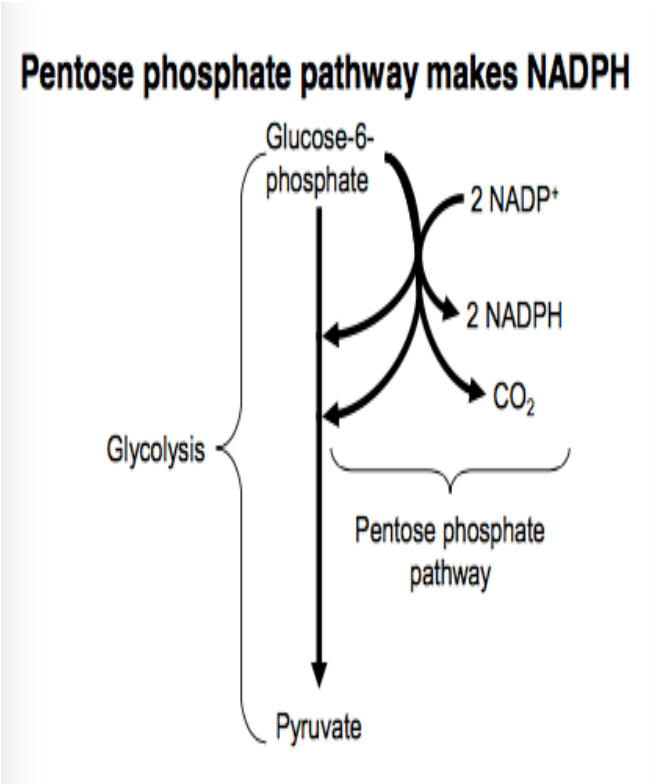
- Excess glucose is stores as glycogen in a process that is regulated allosterically and by enzyme phosphorylation in response to hormones
- In glycolysis, 1 molecule of glucose is oxidized to pyruvate, forming 2ATP and 2 NADH
- Glycolysis is regulated allosterically and by availability of glucose
- Fermentation produce ATP in the absence of oxygen, with no net oxidation of carbon

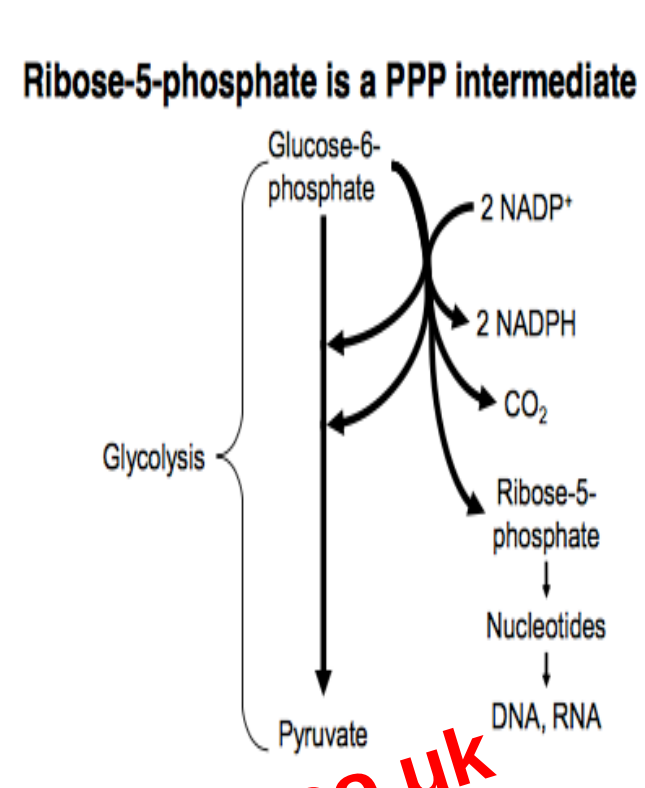
**4. Pentose Phosphate Pathway (PPP)**

- Why does it exist?
    - Main source of NADH
- Under conditions that **cell has a lot of insulin and energy is high**, this pathway is turned on
    - This means that biosynthesis is favoured (favoured when energy status is good)
- Remove electrons from carbon and give it to NADPH
- Depending on conditions of the cell, there are different versions of PPP, sometimes not all the carbons will be diverted back to carbons—sometimes, the carbons will be diverted to ribose-5-phosphate instead
    - Usually shift to this version when we don't need the energy from glycolysis, have energy to spare for biosynthesis
    - Ribose-5-phsphate make up nucleotides which makes up DNA and RNA
- Important for maintenance in the body—restocking on nucleotides and NADH
- However, when energy is low in body, this process turned off

**When its turned on and the ribose-5 phosphate pathway






**Gluconeogenesis and PPP summary:**

- Liver can make glucose when glycogen stores run out, at a cost 6 ATP equivalents per glucose
- Gluconeogenesis is not simply the reverse of glycolysis
- Regulation of gluconeogenesis is opposite to that glycolysis
- Pentose phosphate pathway can produce NADPH and ribose-5- phosphate



**TOPIC 13: pyruvate oxidation and kreb's cycle**

**Mitochondrion:**

- The mitochondrion is made up of many small pores that allow small molecules (less than 5000 Da) to cross it's membrane
- The inner membrane prevents ions and polar molecules from crossing, unless a special pathway exists (i.e. pyruvate oxidation pathway)
- The inner membrane is folder to form cristae
- The matrix is the space enclosed by the inner mitochondrial membrane

Pyruvate Oxidation:

- Occurs after glycolysis in the mitochondrion

$$\text{Pyruvate} + O_2 \longrightarrow CO_2 + H_2O$$

1. Decarboxylation of pyruvate to form acetyl-coenzyme A (acyl-coA)→3 carbon to 2 carbon molecule releasing carbon dioxide
2. Oxidation of the acetyl of acetyl-coA in the citric acid cycle

**know how to recognize the structure of coenzyme A

Coenzyme A: