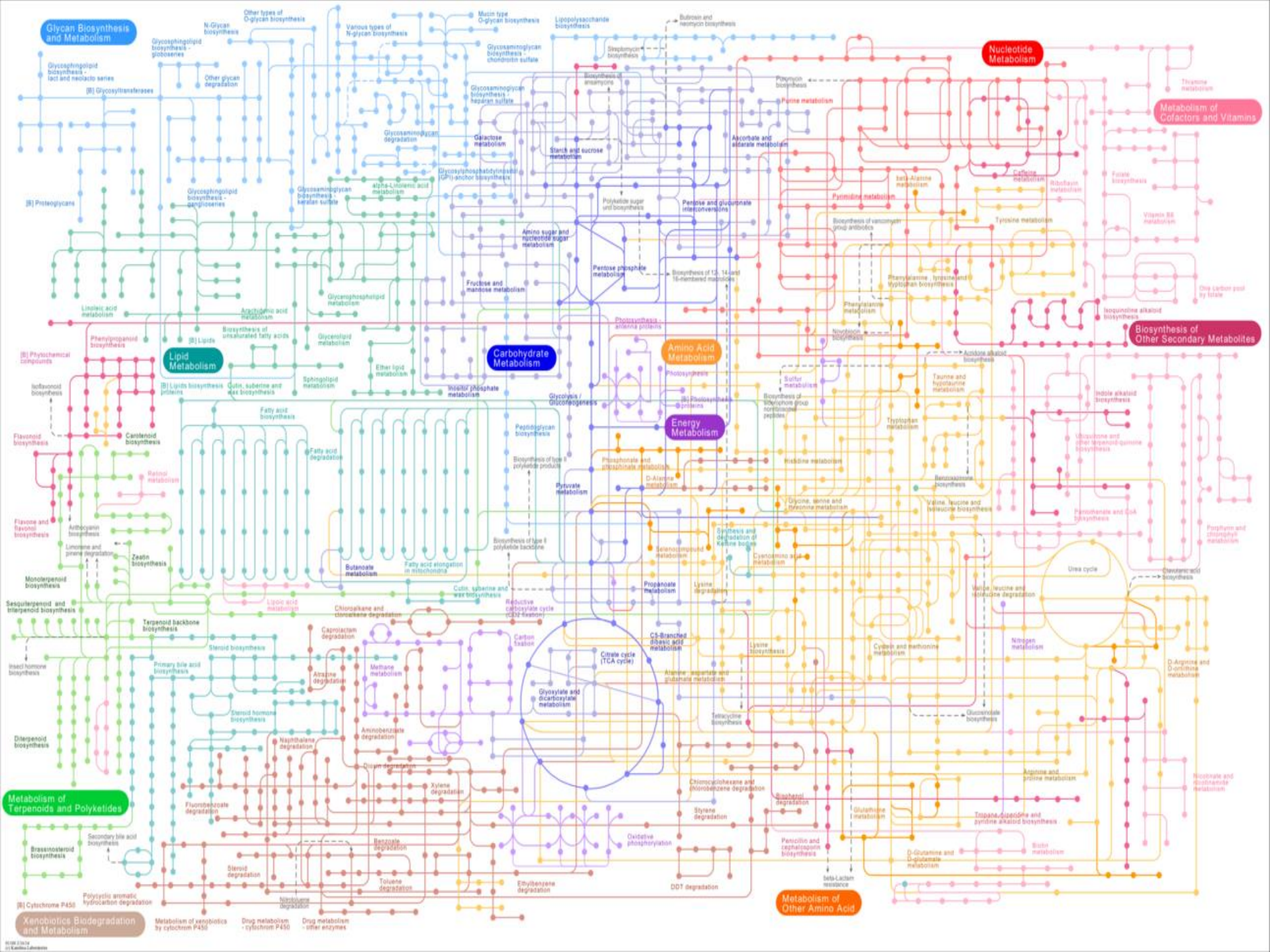


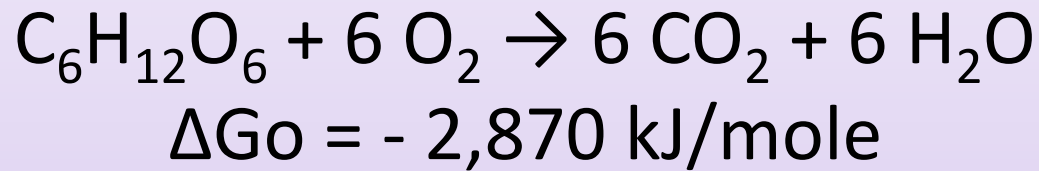
Session 4
Lecture 1
Oxidative Phosphorylation

Dr. Abdullah Ali



Oxidative Phosphorylation (Stage 4 of Metabolism)

- The complete oxidation of glucose can be represented by the following equation:

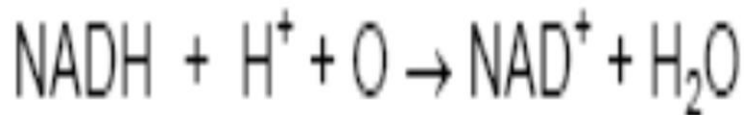
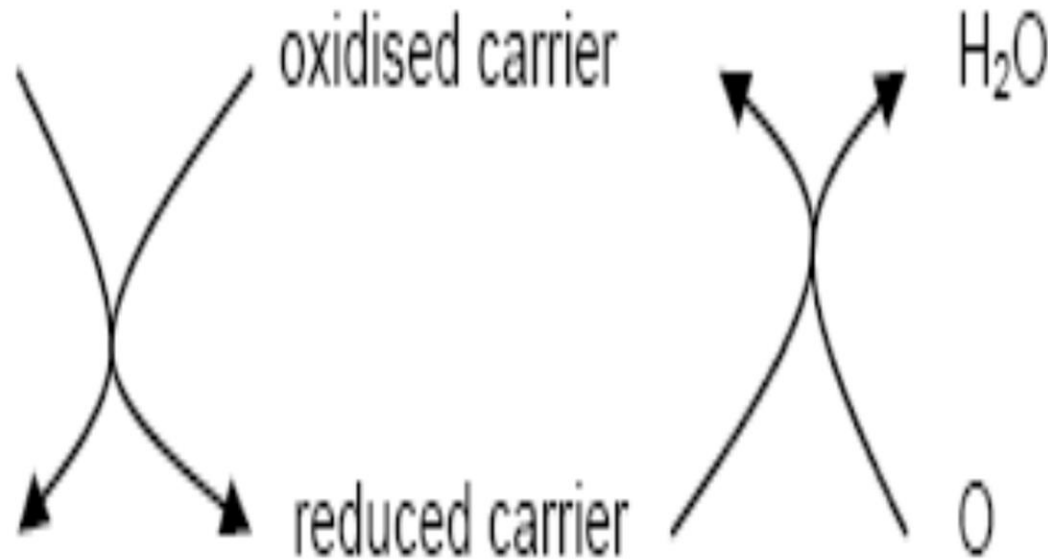


- Till now only 2ATP and 2GTP are formed from oxidation of glucose which equal to 124 kJ/mole
- The question is Where has all the energy gone?
- The answer is that energy is still carried by the carriers of the reducing equivalents (NADH & FADH₂)

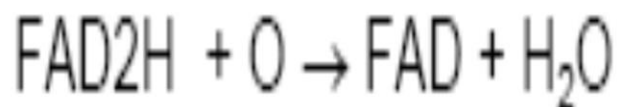
Consider the energy liberated from oxidation of these carriers:

- $\text{NADH} + \text{H}^+$
(FAD2H)

NAD^+
(FAD)



$$\Delta G^\circ = -220 \text{ kJ/mole}$$



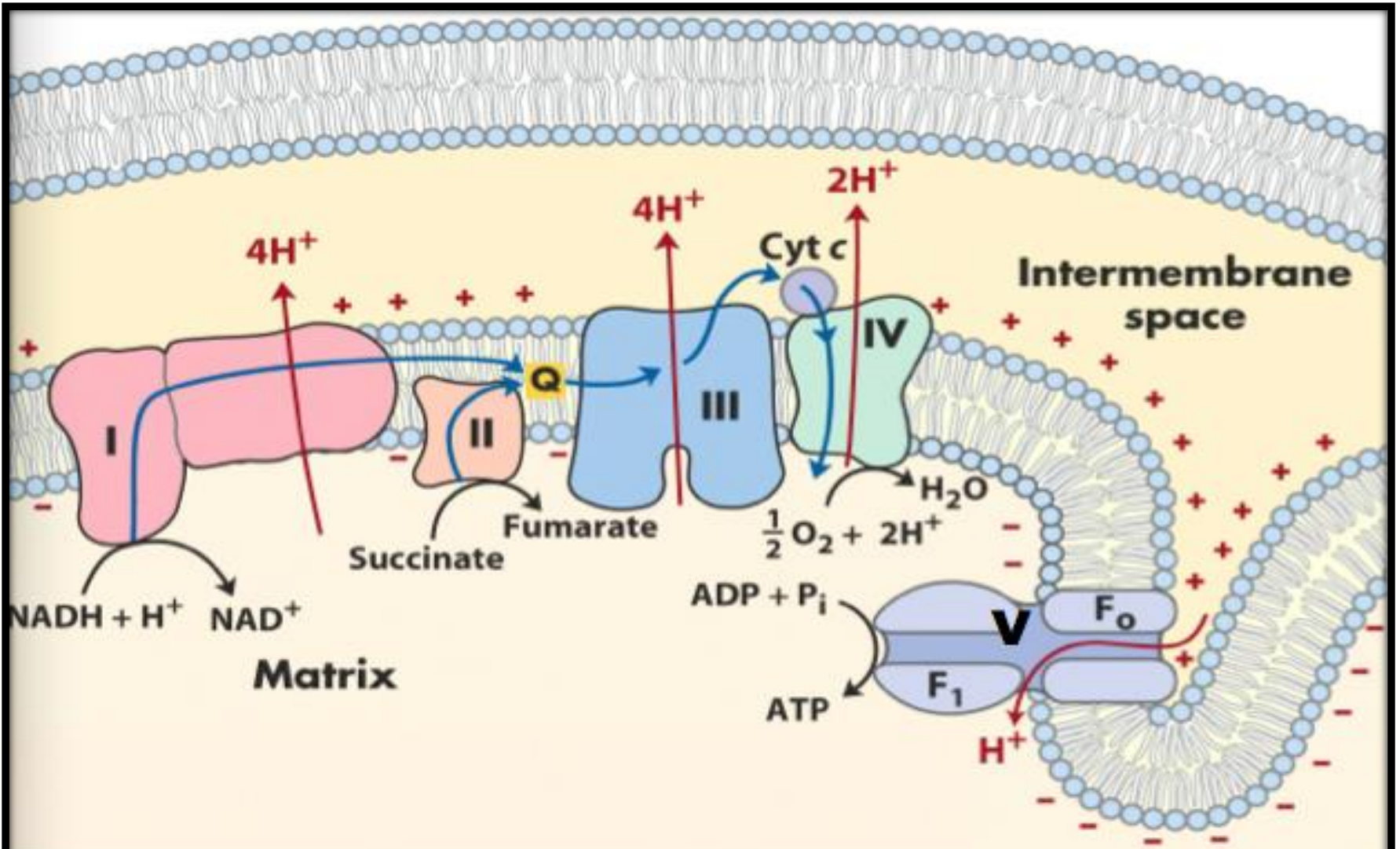
$$\Delta G^\circ = -152 \text{ kJ/mole}$$

Oxidation of NADH and FADH₂

- It is carried out in a pathway called oxidative phosphorylation (OP)
- OP represents 2 processes, the first is the oxidation of NADH and FADH₂ in the **electron transport chain** and the second **phosphorylation** of ADP to produce ATP
- **The energy liberated from the oxidation of the carriers of the reducing equivalents was used for phosphorylation of ADP to produce ATP**

Electron Transport Chain (ET)

- **complex I** (NADH-Q oxidoreductase): oxidizes NADH and transfers electrons to coenzyme Q
- **coenzyme Q** (Ubiquinone)
- **complex II** (Succinate-Q reductase): oxidizes FADH_2 and transfers electrons to coenzyme Q
- **complex III** (Q-cytochrome c oxidoreductase): passes the electrons to cytochrome c
- **complex IV** (Cytochrome c oxidase): passes the electrons to O_2
- **Complex V** (ATP synthase): production of ATP.



Note: Complex II contains succinate dehydrogenase (citric acid cycle)

- Three of the complexes (I, III and IV) in addition to transferring electrons, also act as **proton translocating complexes**.
- They use the **free energy** to move protons from the **inside** to the **outside** of the inner mitochondrial membrane. The membrane itself is impermeable to protons
- The proton translocating complexes transform the chemical bond energy of the electrons into an electro-chemical potential difference of protons. **This is known as the proton motive force (p.m.f).**

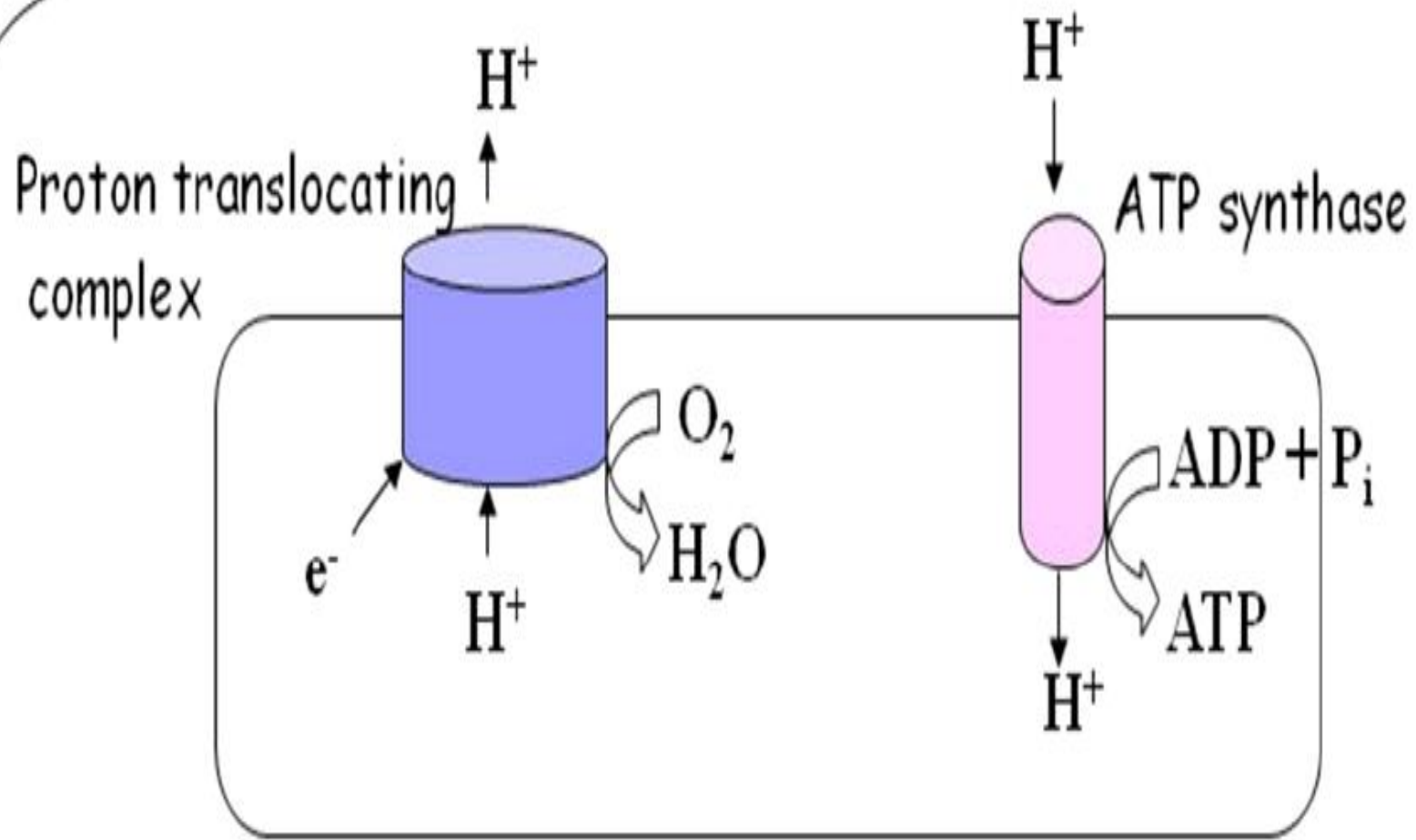
How does ATP synthesize after oxidation of NADH and FADH₂?

- ATP hydrolysis results in the release of energy:



- This means that ATP synthesis from ADP and Pi requires +31 kJ/mole to drive the reaction
- This amount is derived from the **p.m.f** that has been produced across the inner mitochondrial membrane .
- Protons can normally only re-enter the mitochondrial matrix via the ATP synthase complex, driving the synthesis of ATP from ADP and Pi as in the following:





Coupling between electron transport (ET) and ATP synthesis

Normally ET and ATP synthesis are tightly coupled. The mitochondrial concentration of ATP regulates both processes. **When [ATP] is high:**

- The [ADP] is **low** and the ATP synthase stops (lack of substrate).
- This prevents the transport of protons back into the mitochondria.
- The [H⁺] outside increases to a level that prevents more protons being pumped.
- In the absence of proton pumping, the electron transport stops.
- The reverse occurs **when [ATP] is low.**

Inhibitors of OP

- **Uncouplers**
- **Inhibitors of electron transport**
- **Inhibitors of ATP synthase**

Uncouplers

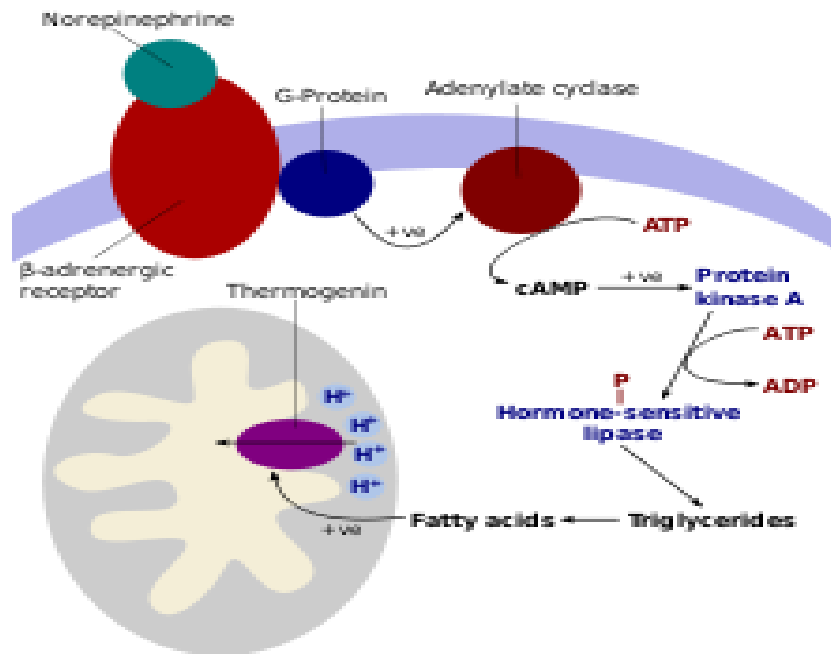
- Some synthetic substances like **dinitrophenol** and **dinitroresol** increase the permeability of the inner mitochondrial membrane to protons
- ET continues, ATP synthesis stops and energy is dissipated as heat (**thermogenesis**).
- **Uncoupling proteins** (UCP1 to UCP5) are identified in tissues to produce heat.

Uncoupling proteins

- UCP1-3 are important to induce a leak of protons across the membrane, **reducing** the **p.m.f** and **inhibiting ATP synthesis**.
- Proton leak is physiologically important and accounts for 20-25% of the **basal metabolic rate**.
- UCP1 (previously known as **thermogenin**) is expressed in brown adipose tissue and is involved in non-shivering **thermogenesis**, which enables mammals to survive in cold environments.

Uncoupler Proteins

- In response to cold, noradrenaline is released and stimulates lipolysis releasing fatty acids to provide fuel for oxidation in brown adipose tissue
- Fatty acids breakdown produce FADH₂ and NADH, driving ET and increasing the **p.m.f.** Noradrenaline also activates UCP1 to inhibit ATP synthesis, which dissipates the **p.m.f.** as heat.
- UCP3 has been found in skeletal muscle, brown adipose tissue and the heart and it appears to be involved in modifying fatty acid metabolism



Inhibitors of Electron Transport

- **Carbon monoxide, cyanide, rotenone and antimycin are inhibitors for components of ET.**
- **Without the p.m.f, ATP cannot be synthesized and no heat is generated. Irreversible cell damage rapidly occurs**

Oxidative stress

- During oxidative phosphorylation, **reactive oxygen species** are produced as a by-product.
- Mitochondria produce superoxide radicals ($O_2^{\cdot -}$) which may be converted to hydrogen peroxide (H_2O_2).
- Some superoxide radicals react with nitric oxide to form peroxynitrite ($ONOO^-$) and hydroxyl radicals ($\cdot OH$).
- These highly reactive agents can cause **damage to cells**, particularly to membranes.
- Some cells, such as **neutrophils** release large amounts of ROS to destroy bacteria