Bioenergetics & Biological oxidation

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Bioenergetics

Definition

''Bioenergetics or biochemical thermodynamics is the study of energy changes in biochemical reactions.''

Non biologic systems use heat energy to accomplish work but biologic systems are isothermic & utilize chemical energy for the living process.

Bioenergetics

Laws of thermodynamics

1- First Law:

"The Total energy of a system is constant , including its surroundings."
This is also the law of conservation of energy.
Energy may be transferred from one part to another or may be transformed into an other form of energy.

2- **Second Law:**

"The total entropy of a system must increase if a process is to occur spontaneously."
Entropy represents the extent of disorder of the system & becomes maximum when it approaches true equilibrium.

Bioenergetics

Free Energy

It is useful energy also known as the chemical potential.

Gibbs change in free energy (ΔG) is that portion of the total energy change in a system available for doing work.
Under constant temperature & pressure, the relationship between the free energy change (ΔG) & the change in entropy (ΔS) =

$\Delta G = \Delta H - T\Delta S$.

ΔH = Enthalpy (Energy content)

T = the absolute temperature

Entropy- Randomness of the system

Bioenergetics

Types of reactions

Exergonic reaction: It is a spontaneous reaction that releases energy. If the free energy change is negative, this reaction is due to loss of energy from reactants, so it is called exergonic.
E.g. catabolic reactions.
Endergonic reaction: It is an anabolic reaction that consumes energy. If the free energy change is positive, the reaction is called endergonic. E.g. synthetic reactions,
But at equilibrium, it is zero
The energy coupling occurs by coupling of Exergonic and endergonic reactions and liberation of heat.

Bioenergetics

Energy currency of the cell

ATP is the primary and universal carrier of chemical energy in the cell.
Terminal (alpha) phosphate group of ATP on hydrolysis yields - 7.3 kcl/mol

ATP=ADP + Pi=7.3 kcal/mol

Bioenergetics

Phosphagens

They are storage forms of high energy phosphates.

Eg: Creatine phosphate in vertebrate muscle & brain,

Arginine phosphate in invertebrate muscle.

In physiologic conditions , phosphagens permit ATP conc. to be maintained in muscle when ATP is used as a source of energy for muscle contraction.

When ATP is abundant, its contraction can cause the reverse reaction to take place & allow the conc. of creatine phosphate to increase abundantly so as to act as a store of creatine phosphate.

Bioenergetics

Interconversion of Adenine Nucleotides

The enzyme Adenylate Kinase (Myokinase) is present in most cells.
It catalyzes the interconversion of ATP & AMP on the one hand and ADP of the other.
The reaction has three functions-
1-It allows high energy phosphate in ADP to be used in the synthesis of ATP.
2-It allows AMP to be recovered by rephosphorylation to ADP.
3-It allows AMP to increase in conc. When ATP becomes depleted and act as a metabolic signal to increase the rate of catabolic reactions which, in turn leads to the generation of more ATP.

Bioenergetics

Nucleoside phosphates related to ATP & ADP

Nucleoside triphosphates similar to ATP but containing an alternative base to adenine can be synthesized from their diphosphates by means of the enzyme nucleoside diphosphate kinase, e.g.:

ATP + UDP ⟷ (Nucleoside diphosphate kinase) ADP + UTP

ATP + GDP ⟷ ATP + GTP

ATP + CDP ⟷ ADP+ CTP

All these triphosphates take part in phosphorylations in the cell. Similarly , nucleoside monophosphate kinases catalyse the formation of nucleoside diphosphates from the corresponding monophoshates.

Biological oxidation

Biological oxidation is the cellular process in which the organic substances release energy (ATP), produce CO2 and H2O through oxidative-reductive reactions.

Organic substances: carbohydrate, fat and protein

Biological oxidation

Oxidation

Chemically defined as removal of electrons

Reduction

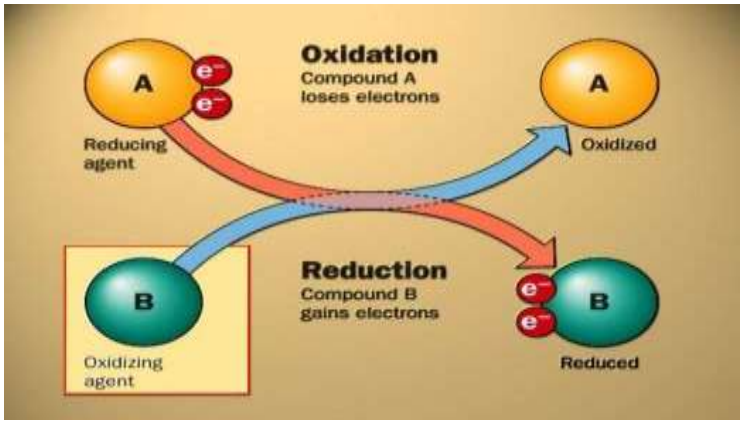
It is defined as the addition of electrons.

E.g.- Fe++ is oxidixed to Fe+++ removal of electron

Fe+++ is reduced to Fe++ electron added

Redox Reaction

The reactions which involves both reduction process & complementary oxidation process called redox reaction.



Biological oxidation

Redox Potential

In oxidation & reduction reactions, the free energy exchange is proportionate to the tendency of reactants to donate or accept electrons. This is expressed as an Oxidation-reduction or redox potential.

Biological oxidation

Redox Couple

A biological system which has a strong tendency to donate electrons has a negative redox potential

The redox potential of a system is usually compared against the potential of hydrogen electrode at pH 7. 0 with -0.42 volt in biological system.

E.g.	Redox Potential	Redox Pair
	-0.32	NADH/NAD+
	+0.82	H2/1/2O2

The electrons flow from electro negative redox couple to more electro positive system

Biological oxidation

Enzymes & coenzymes involved in Oxidation & Reduction

In the year 1961, the International Union of Biochemistry has designated all enzymes concerned in oxidative processes and oxidoreductases. These are classified as:

1-Oxidases
2-Aerobic Dehydrogenases
3-Anaerobic Dehydrogenases
4-Hydroperoxidases
5-Oxygenases

Biological oxidation

1-Oxidases

Enzymes that catalyse the removal of hydrogen from a substrate but use only oxygen as a hydrogen accepter to form water as a reaction product(with the exception of uricase & monamine oxidase which form H2O2.

A-Cytochrome oxidase

B-Phenolase (Tyrosinase, Polyphenol oxidase, Catechol Oxidase)

C-Laccase

D-Ascorbic Oxidase

E-Uricase

F-Monoamine Oxidase

Biological oxidation

2-Aerobic Dehydrogenases

They catalyze the removal of hydrogen from a substrate and use either oxygen or artificial substances such a methylene blue as hydrogen accepter.

H2O2 is formed as a product.

They are flavoprotein enzymes having FMN(Flavin Mononucleotide) or FAD(Flavin adenine dinucleotide) as prosthetic groups.

A-D-Amino acid dehydrogenase

B-L-Amino acid dehydrogenase

C-Xanthine dehydrogenase (Xanthine Oxidase)

D-Aldehyde dehydrogenase(Aldehyde oxidase)

E-Glucose Oxidase

Biological oxidation

3-Anaerobic Dehydrogenases

They catalyze the removal of hydrogen from a substrate but not able to use oxygen as hydrogen accepter. They transfer hydrogen from one substrate to another by oxidation-reduction reaction not involving a respiratory chain.

They perform oxidation of metabolite utilizing several components of a respiratory chain.

A-Dehydrogenase dependent on Nicotinamide Coenzymes

B-Dehydrogenase dependent on Riboflavin Prosthetic groups

C-Cytochromes-Cytochrome C & Cytochrome 450

Biological oxidation

4-Hydroperoxidases

They utilize hydrogen peroxide as a substrate.

A-Peroxidases

B-Catalase

5-Oxygenases

They catalyse the incorporation of oxygen in to a substrate molecule.

A-Dioxygenase(Oxygen transferases , true oxygenase)

B-Mno-oxygenase(Mixed function oxidases, Hydroxylases)

Biological oxidation

Superoxide dismutase (SOD)

Superoxide dismutase protects aerobic organisms against oxygen toxicity due to superoxide anion free radical ($O^{-2\cdot}$)

$$O_2^{\cdot -} + O_2^{\cdot -} + 2H^+ = H_2O_2 + O_2$$

Catalase removes H_2O_2

Biological oxidation

Respiratory Chain or Electron Transfer Chain

It is the sequence of enzymes and carriers responsible for the transfer of reducing equivalents from substrates to molecular oxygen.

It is located within the Mitochondria.

The components of respiratory chain are arranged in order of increasing redox potential i.e. from NAD+ / NADH to O_2/H_2O redox couples

Components of respiratory chain:

Complex I : NADH : ubiquinone oxidoreductase

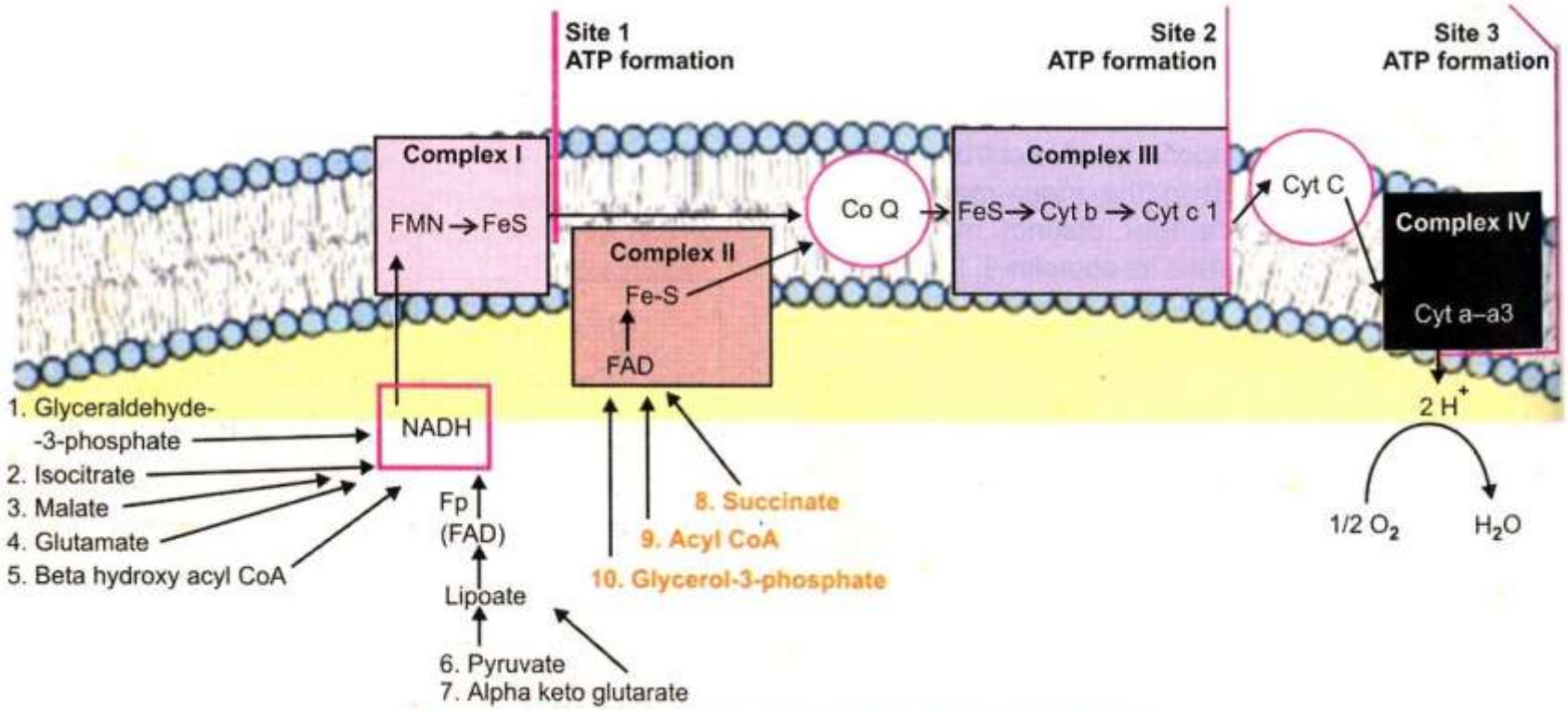
Complex II : succinate : ubiquinone oxidoreductase

Complex III : Ubiquinol : cytochrome C oxidoreductase

Complex IV : cytochrome a,a3: oxygen oxidoreductase

Biological oxidation

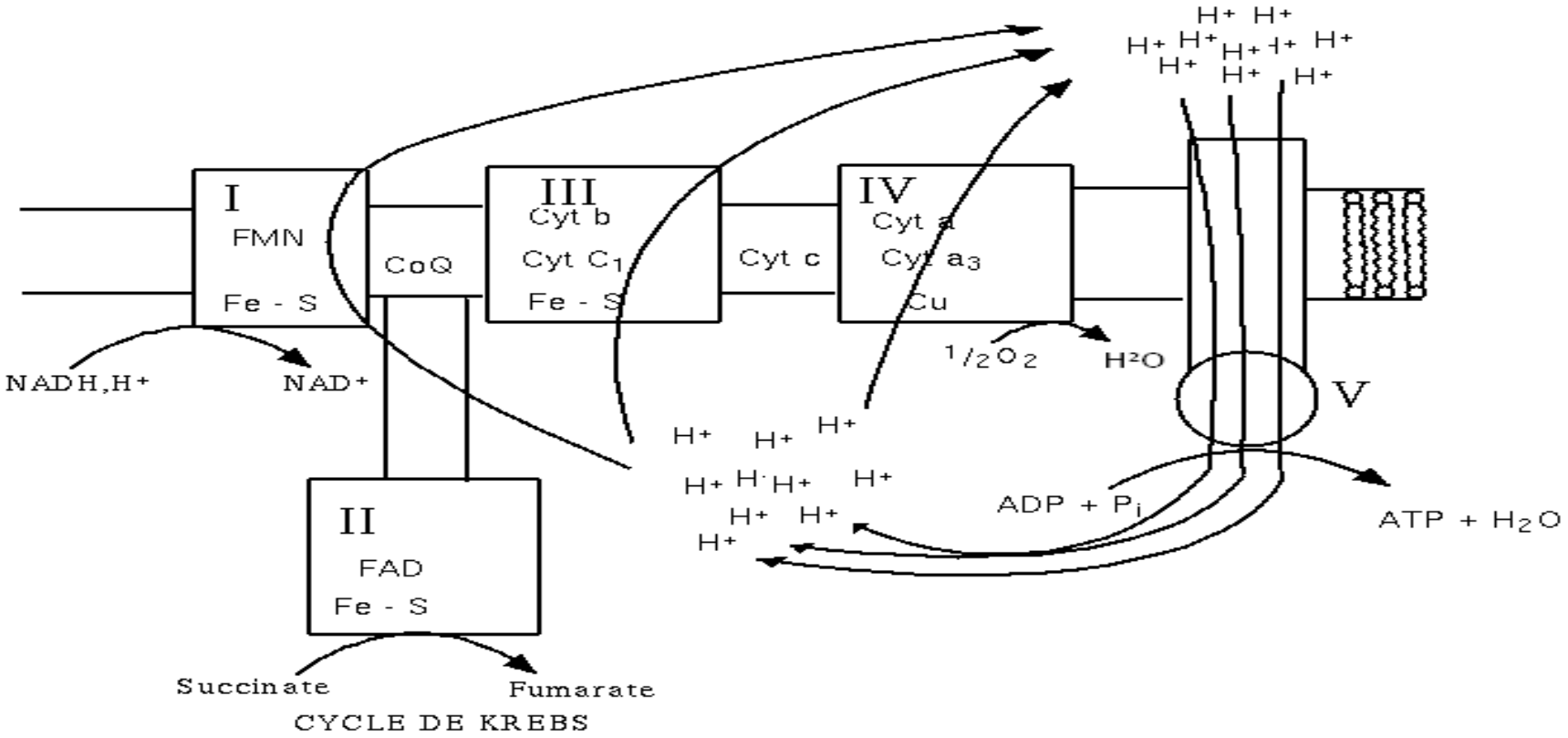
Respiratory Chain or Electron Transfer Chain



Components and sequence of reactions of electron transport chain

Biological oxidation

Respiratory Chain or Electron Transfer Chain



Biological oxidation

Respiratory Chain or Electron Transfer Chain

- Iron sulphur centres (Fe-S) form prosthetic groups of iron sulphur proteins
- Non heme proteins also function in the transfer of electrons e.g. from $FMNH_2$ to Coenzyme Q and cytochrome b to cytochrome c_1.
- Coenzyme Q_{10} is a fat soluble quinone compound.
- Cytochromes b,c,c_1, a,a_3 containhaem.
- Cytochromes a,a3 also contains copper.
- ATP synthetase is involved in the formation of ATP.

Biological oxidation

Respiratory Chain or Electron Transfer Chain

Summary of Electron flow in ETC

Complex I: NADH → FMN → Fe-S → Co Q →

Complex II: Succinate → FAD → Fe-S → Co Q →

Complex III: Co Q → Fe-S → cyt.b → cyt.c1 → cyt. c

Complex IV: Cyt. c → cyt a-a3 → O_2

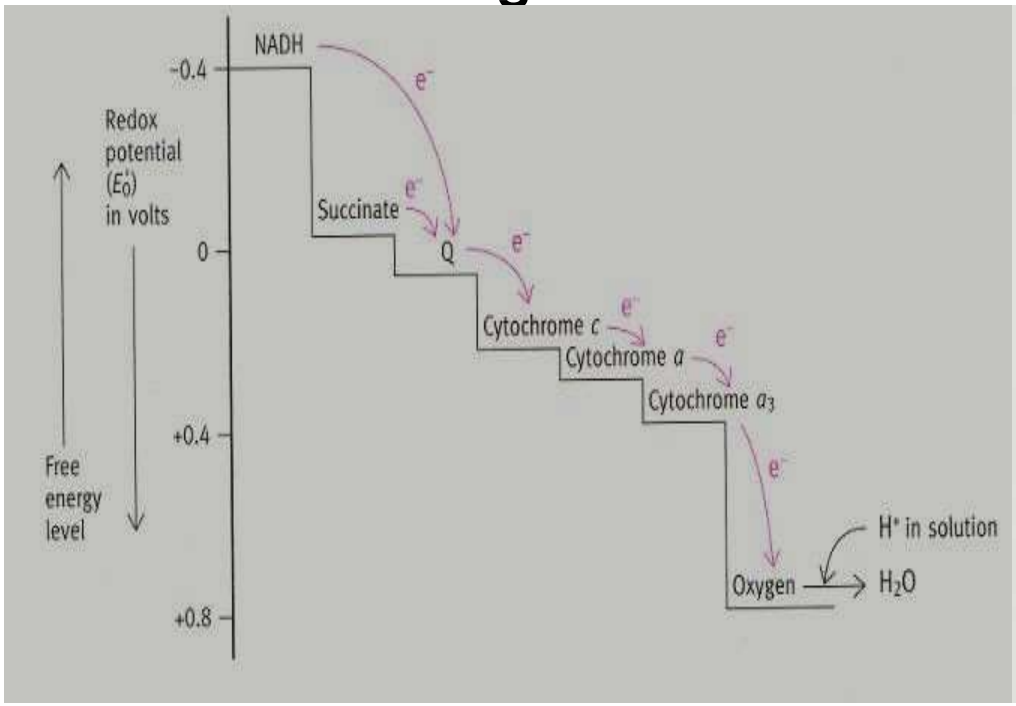
Protein components of Mitochondrial Electron Transfer Chain

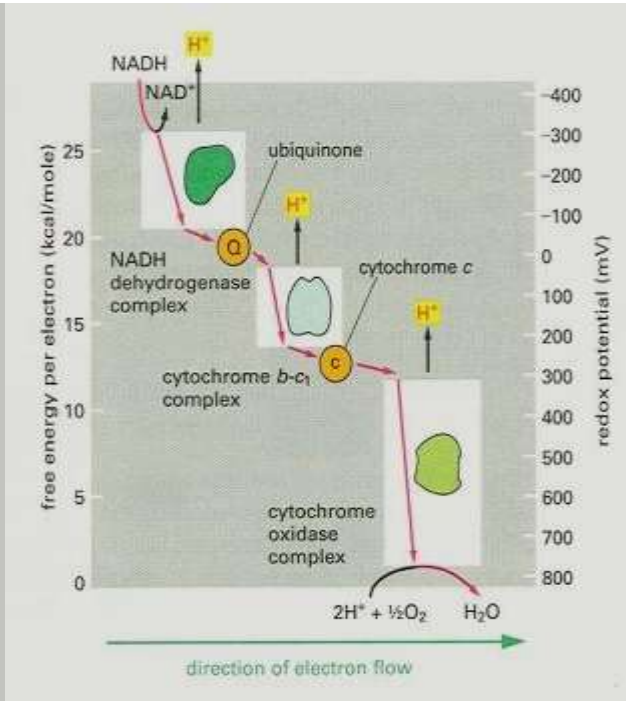
Enzyme complex/protein	Mass (kDa)	Number of subunits*	Prosthetic group(s)
I NADH dehydrogenase	850	43 (14)	FMN, Fe-S
II Succinate dehydrogenase	140	4	FAD, Fe-S
III Ubiquinone:cytochrome *c* oxidoreductase	250	11	Hemes, Fe-S
Cytochrome $c^{\dagger}$	13	1	Heme
IV Cytochrome oxidase	160	13 (3–4)	Hemes; Cu_A, Cu_B

Biological oxidation

Respiratory Chain or Electron Transfer Chain

Organization of carriers in ETC





Biological oxidation

Respiratory Chain or Electron Transfer Chain

Inhibitors:

Site-I (Complex-I)
Rotenone: A fish poison and also insecticide. Inhibits transfer of electrons through complex-I-NADH-Q-reductase.

Amobarbital (Amytal) and Secobarbital: Inhibits electron transfer through NADH-Q reductase.

Piericidin A: An antibiotic. Blocks electron transfer by competing with CoQ.

Drugs: Chlorpromazine and hypotensive drug like guanethidine.

Biological oxidation

Respiratory Chain or Electron Transfer Chain

Inhibitors:

Site-II (Complex-III)

- Antimycin A
- BAL (Dimer-Caprol)

These blocks electron transfer from cyt. b to c1

- Hypoglycaemic drugs Phenformin

Site-III(Complex-IV)

- Cyanide, H_2S & Azide Inhibits terminal transfer of electrons to molecular O2.
- CO (Carbon monoxide): Inhibits Cyt. oxidase by combining with O_2 binding site.

Biological oxidation

Oxidative Phosphorylation

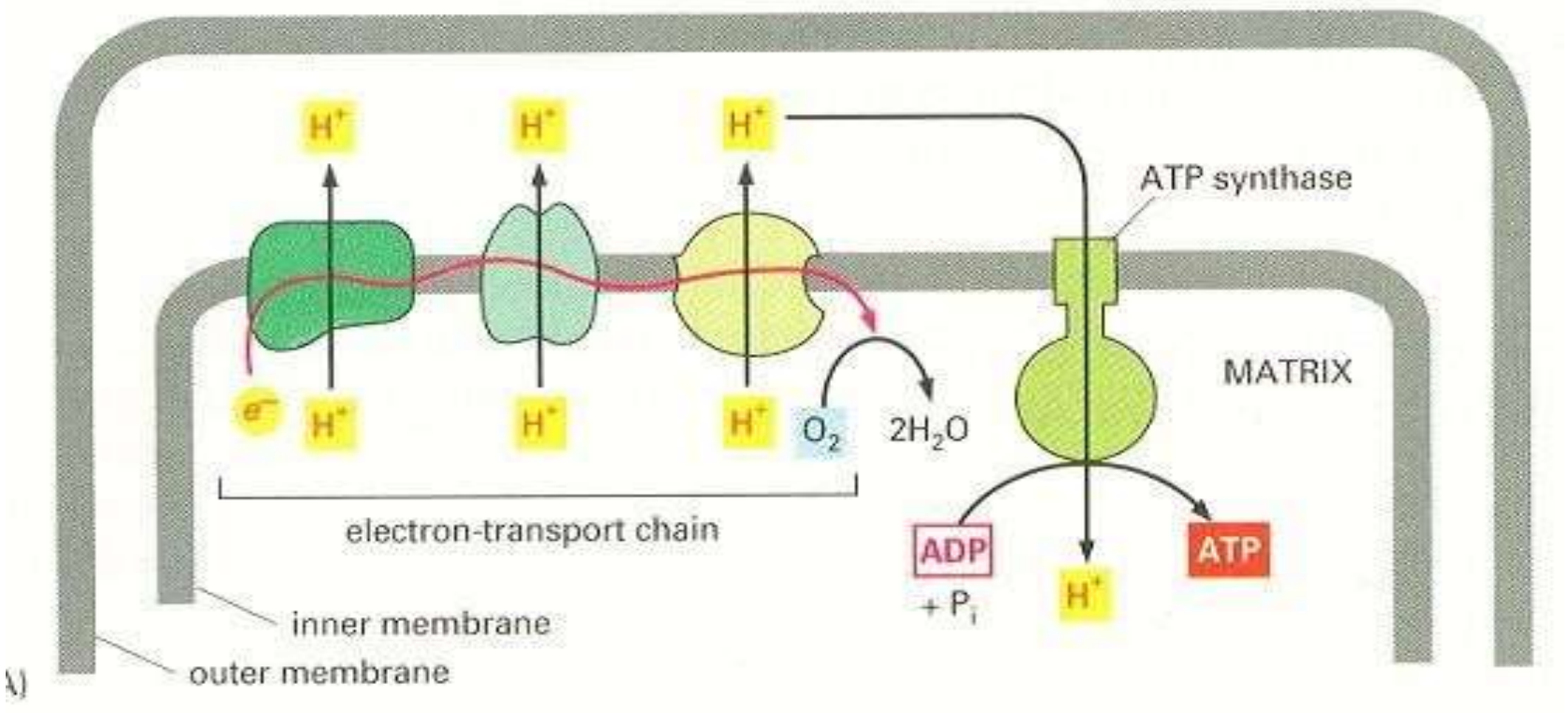
The process by which ADP is phosphorylated by Pi to ATP in the respiratory chain is called Oxidative Phosphorylation.
This process occurs in Mitochondria only.

Chemiosmotic hypothesis

- Proposed by Mitchel
- Dependent on proton gradient.
- According to Mitchell the primary event in oxidative phosphorylation is the translocation of Protons(H+)to the exterior of a coupling membrane driven by oxidation in the Respiratory Chain.
- The membrane is impermeable to protons which accumulate outside the membrane creating an electrochemical potential difference across the membranes. This electrochemical potential difference is utilized to drive a membrane located ATP Synthetase which, in the presence of Pi+ADP, forms ATP.

Biological oxidation

Chemiosmotic hypothesis



Biological oxidation

Oxidative Phosphorylation

P/O Ratio

It is the number of high energy phosphates produced per atom of oxygen used ie : for $NADH+H^+$ - 2.5 ATP

For $FADH_2$ – 1.5 ATP

Respiratory control:

In the presence of adequate O_2 and substrate, ADP becomes rate limiting.

Biological oxidation

Oxidative Phosphorylation

Uncoupling Agents

Uncouplers are the substances that allow electron transport chain to function without phosphorylation and so ATP is not synthesized but oxidation proceeds.

They are lipophilic and allow transport of H^+ across the inner membrane but not through ATP synthetase and so the proton gradient is cancelled without ATP formation and the free energy is liberated as heat e.g.

- 2,4 dinitrophenol(DNP)-it uncouples phosphorylation by the hydrolysis of x~I or X~Pi.
- thermogenin (natural) – uncoupling protein
- Thyroxine-It causes swelling of the mitochondria
- Methylene Blue, Arsenite, Dicoumarol, Aureomycin, & Gramicidin

Biological oxidation

Oxidative Phosphorylation

Inhibitors

Inhibitors completely blocks oxidation & phosphorylation in intact mitochondria

- **Atractyloside** inhibits oxidative phosphorylation. It inhibits the transportation of ADP into the mitochondria and of ATP out of the mitochondria.
- **Ionophores**:
  - An ionophore is a chemical species that reversibly binds ions. Many ionophores are lipid-soluble entities that transport ions across a cell membrane.
  - Ionophore means "ion carrier" as these compounds catalyze ion transport across hydrophobic membranes such as liquid polymeric membranes (carrier-based ion selective electrodes) or lipid bilayers found in the living cells or synthetic vesicles (liposomes)e.g. Valinomycin, Nigercin
  - These alters the permeability of Mitochondrial Membrane.

Biological oxidation

Substrate level phosphorylation

The formation of ATP within certain steps of metabolic pathway i.e. susbstrate level without passing through ETC is called as substrate level phosphorylation

e.g. pyruvate kinase, phosphoglycerate kinase, succinate thiokinase

Biological oxidation

Mitochondrial Diseases

MELAS (Mitochondrial encephalopathy, Lactic acidosis, stroke due to Complex I deficiency
Fatal infantile mitochondrial myopathy and renal dysfunction
Leber's hereditary optic neuropathy
Alzheimer's disease
Parkinsons' disease
Diabetes mellitus


Thank you!