Electron Transport Chain and Oxidative Phosphorylation

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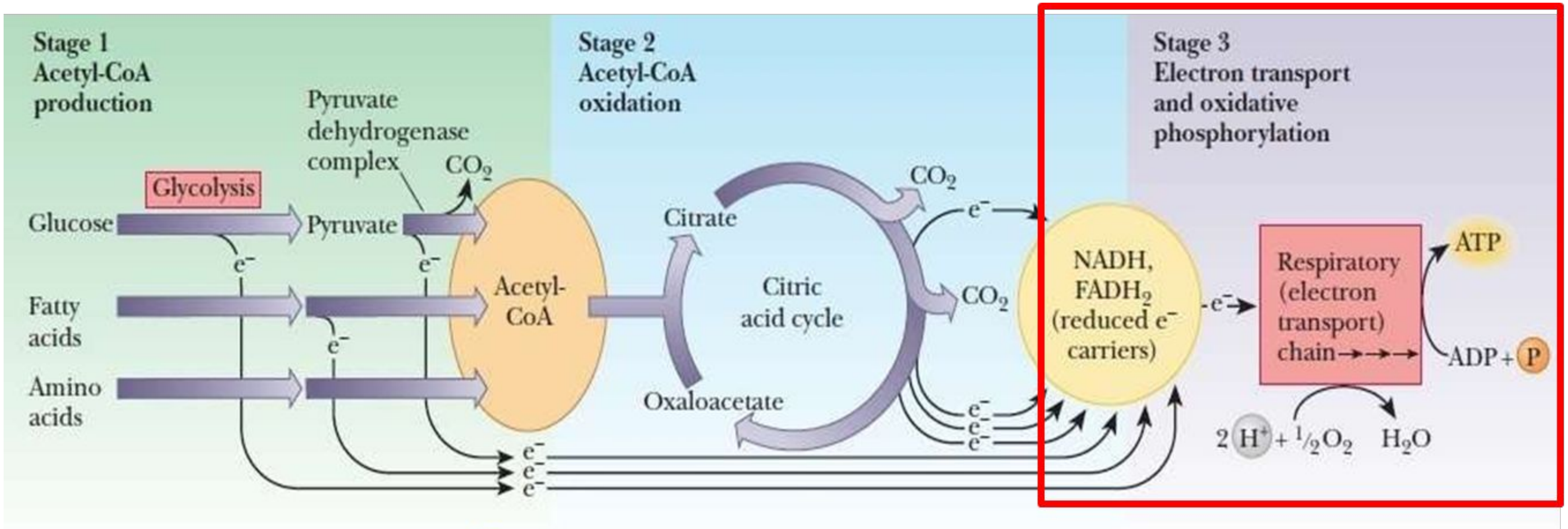
Cellular Respiration



In Cytosol

In Mitochondria

Net Reaction for cellular respiration (for glucose)

$$C_6H_{12}O_6 + 6O_2 + 38ADP + 38P_i \rightarrow 6CO_2 + 6H_2O + 38ATP$$

Electron Transport Chain (Respiratory Chain)

- Is the assembly of respiratory enzymes or carrier proteins found in cristae or inner membrane of mitochondria in eukaryotic cell and plasma membrane of prokaryotic cell.

- **Occurs within all cells except RBCs and the cornea & lens of eyes**
- **Represents the final stage in the oxidation of carbohydrates, fats, and amino acids**
- **Transfers reducing equivalents from NADH & FADH2 to O_2**
- **Primes the process of ATP generation via oxidative phosphorylation**

- **Consists of four large protein complexes (I-IV)**

- **Most active when there is an increased need for ATP**

- The different components of electron transport chain are NAD, FAD, Co-Q, Cyt-b, Cyt.-c1, Cyt.-c2 Cyt.-a, Cyt.-a3.
- NAD and FAD are nucleotide and hydrogen acceptors. Co-Q is the complex organic compound and hydrogen acceptors.
- The mitochondrial respiratory chain consists of a series of sequentially acting electron carriers, most of which are integral proteins with prosthetic groups capable of accepting and donating either one or two electrons.
- Three types of electron transfers occur in oxidative phosphorylation:
  - (1) direct transfer of electrons, as in the reduction of Fe3 to Fe2;
  - (2) transfer as a hydrogen atom (H *e*); and
  - (3) transfer as a hydride ion (**:**H), which bears two electrons.

The term **reducing equivalent** is used to designate a single electron equivalent transferred in an oxidation-reduction reaction.

In addition to NAD and flavoproteins, three other types of electron-carrying molecules function in the respiratory chain:

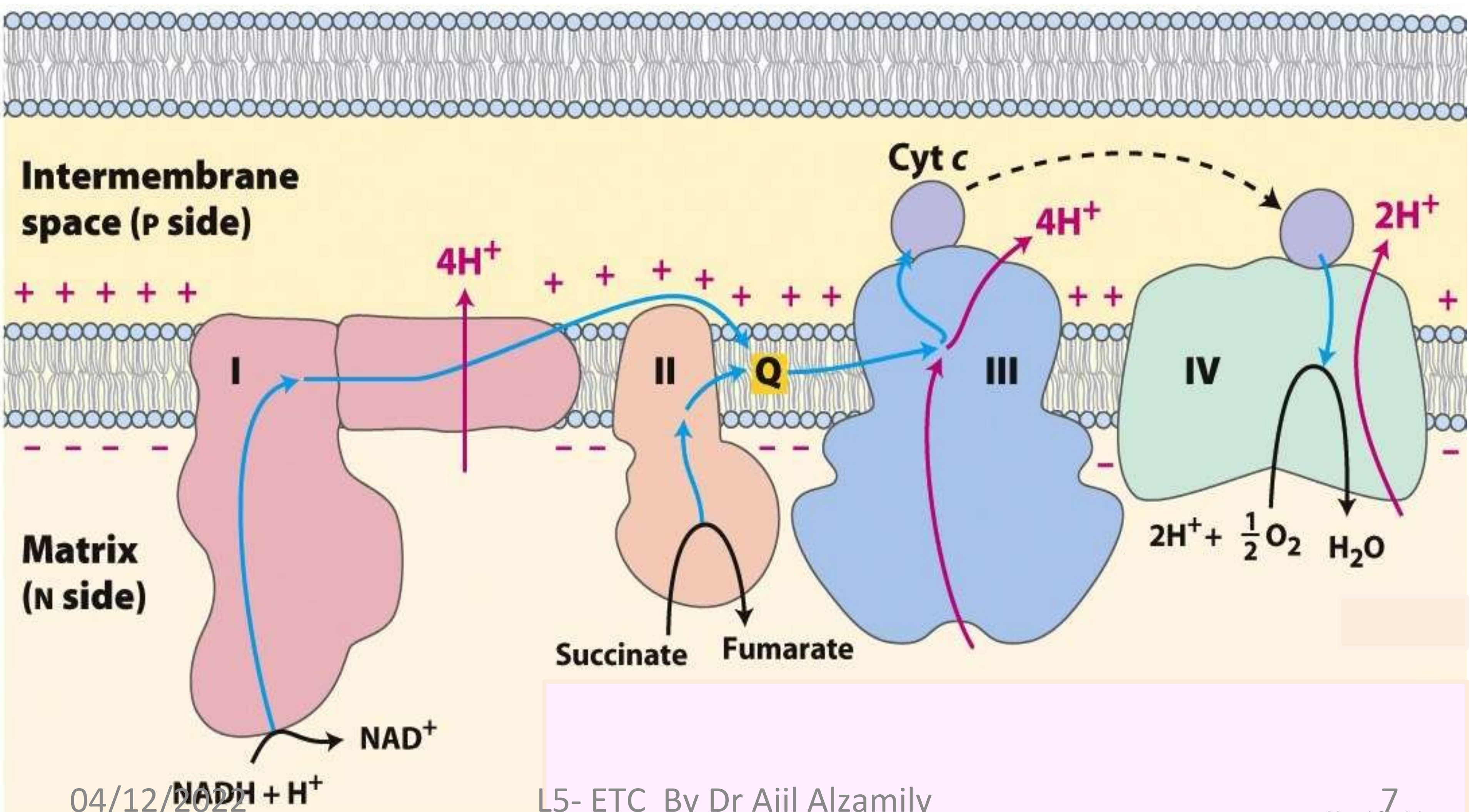
- a hydrophobic quinone (ubiquinone) and
- two different types of iron-containing proteins (cytochromes and iron-sulfur proteins).

- **Ubiquinone (also** called **coenzyme Q, or simply Q) is a lipid-soluble benzoquinone**

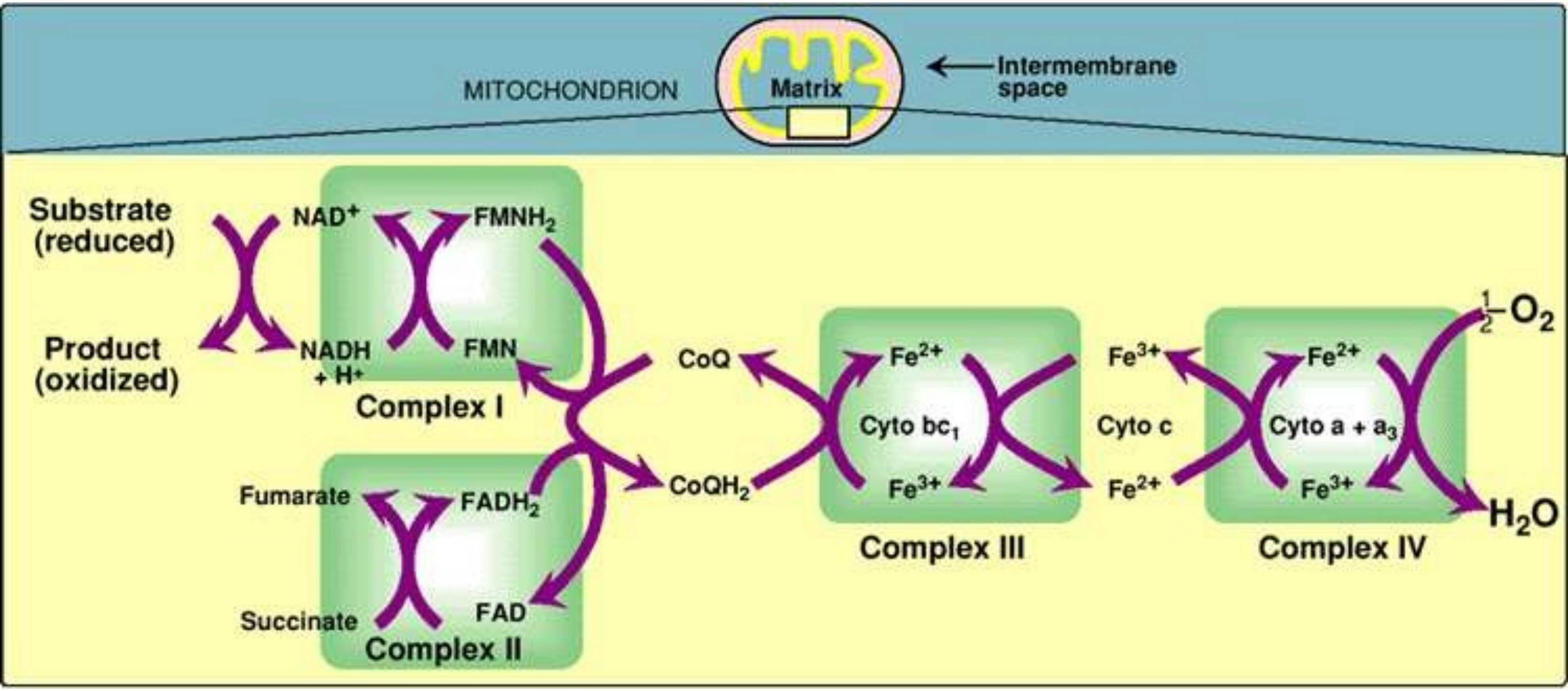
Components of Electron Transport Chain

Complex	Name	No. of Amino acids	Prosthetic Groups
Complex I	NADH-CoQ Reductase	43	FMN, 9 Fe-S cntrs.
Complex II	Succinate-CoQ Reductase	4	FAD, cyt b_{560}, 3 Fe-S centres.
Complex III	CoQ-cyt c Reductase	11	cyt b_H, cyt b_L, cyt c_1, $^{Fe-S}$Rieske
Complex IV	Cytochrome Oxidase	13	cyt a, cyt a_3, Cu_A, Cu_B

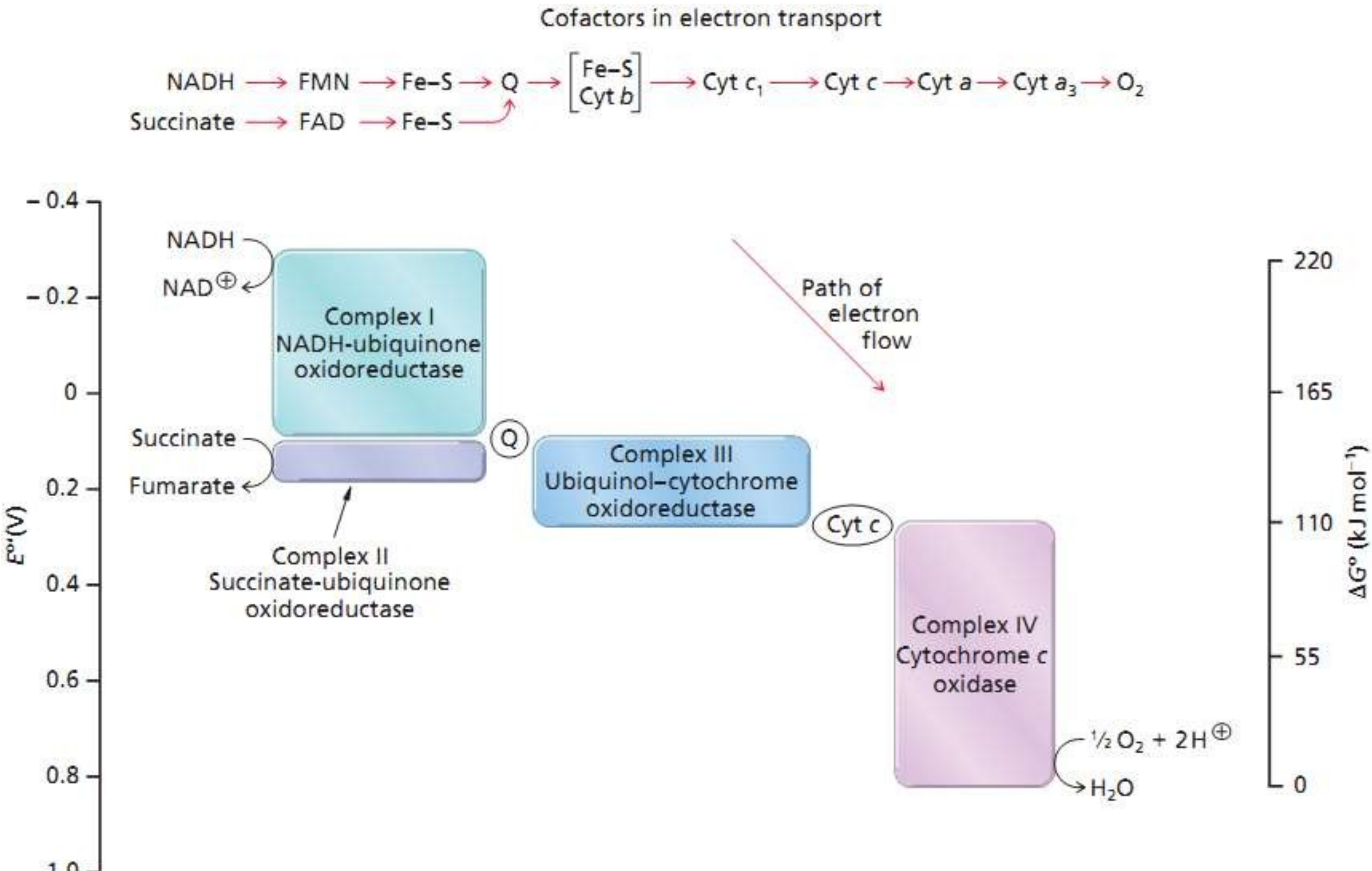
Organization of the Electron Transport Chain



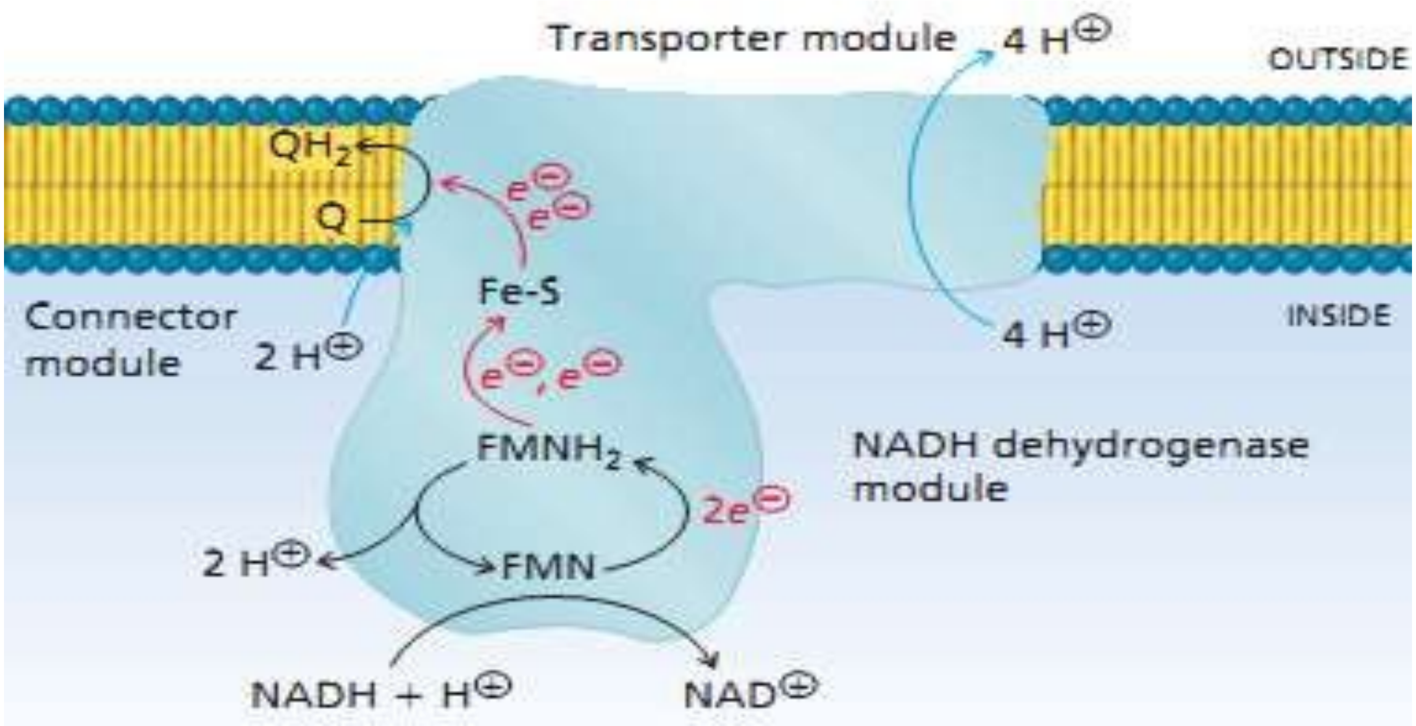
Schematic Representation of ETC



The Sequence of Electrons Transport

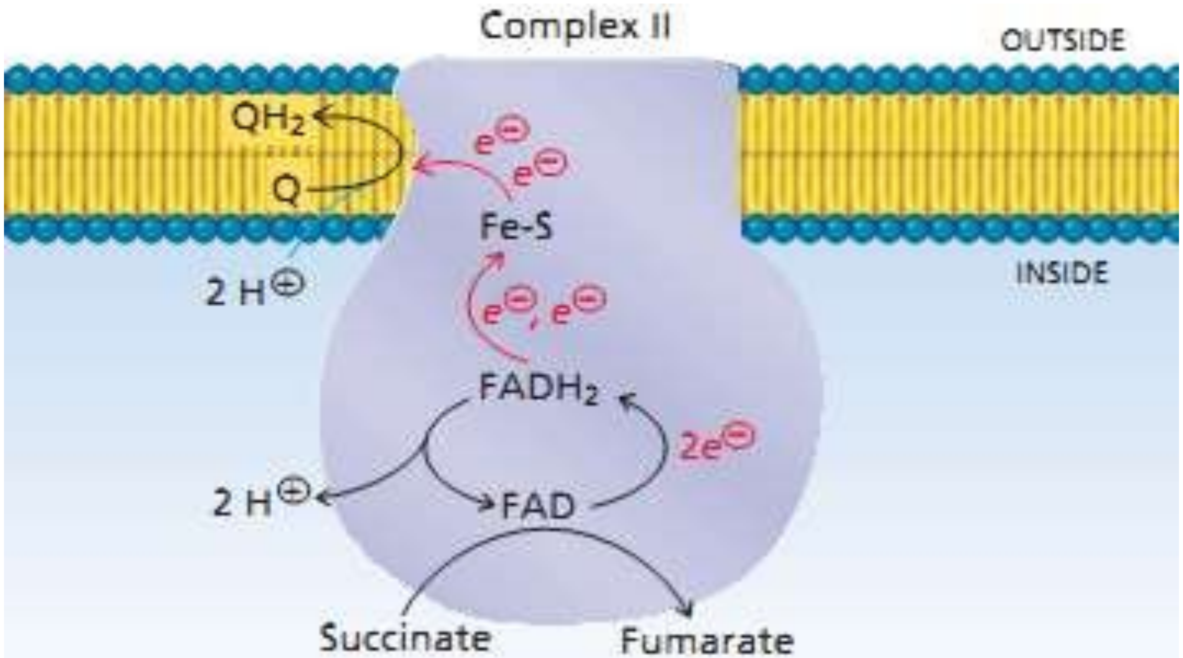


Complex I

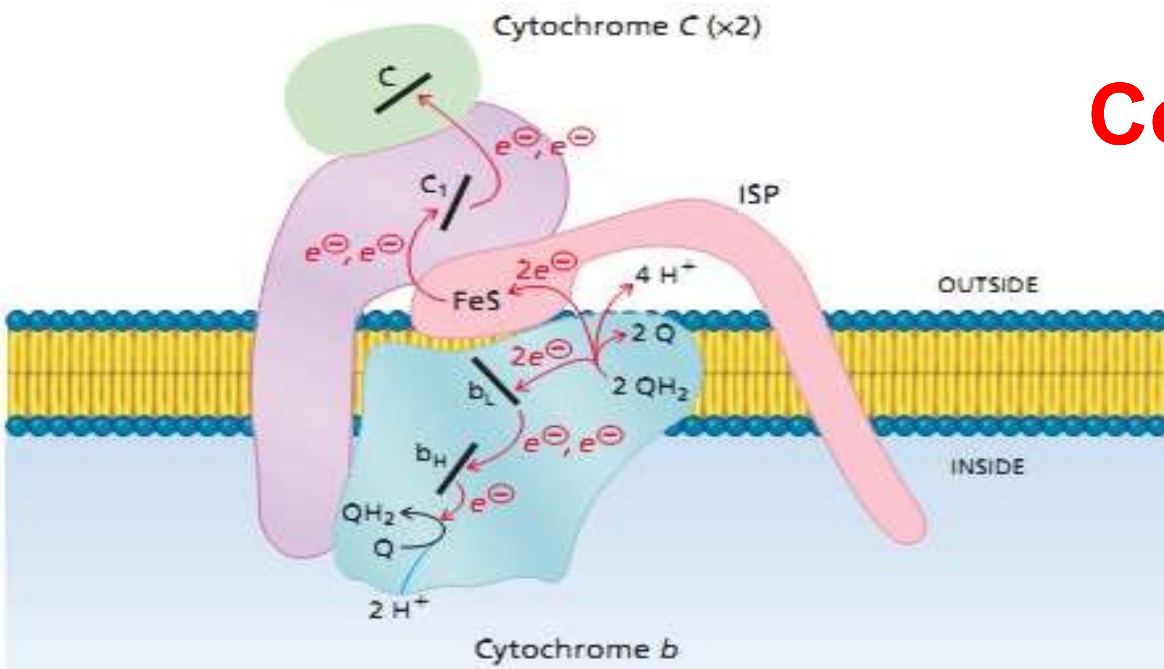


- Aka NADH-Ubiquinone Oxidoreductase
- Contains FMN and Fe-S proteins as the prosthetic groups
- Oxidizes NADH and reduces coenzyme Q:
- Passes electrons to complex III via coenzyme Q
- Transports $4H^+$ ions from the matrix to the cytosol

Complex II



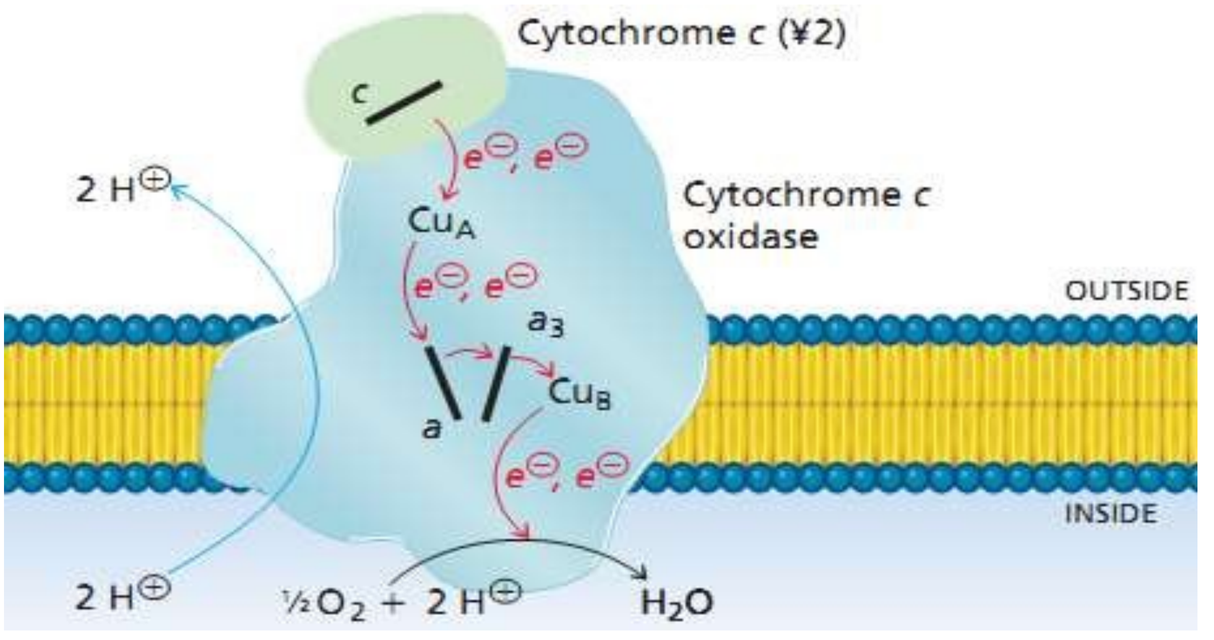
- **Aka succinate-ubiqunione oxidoreductase**
- **Contains FAD, Fe-S proteins and Q as the electron carriers**
- **Unlike other complexes, does not transport H^+ ions**
- **Accepts electrons from succinate and then transfer it to coenzyme Q**
- **Also catalyses one of the reactions of TCA cycle**



Complex III

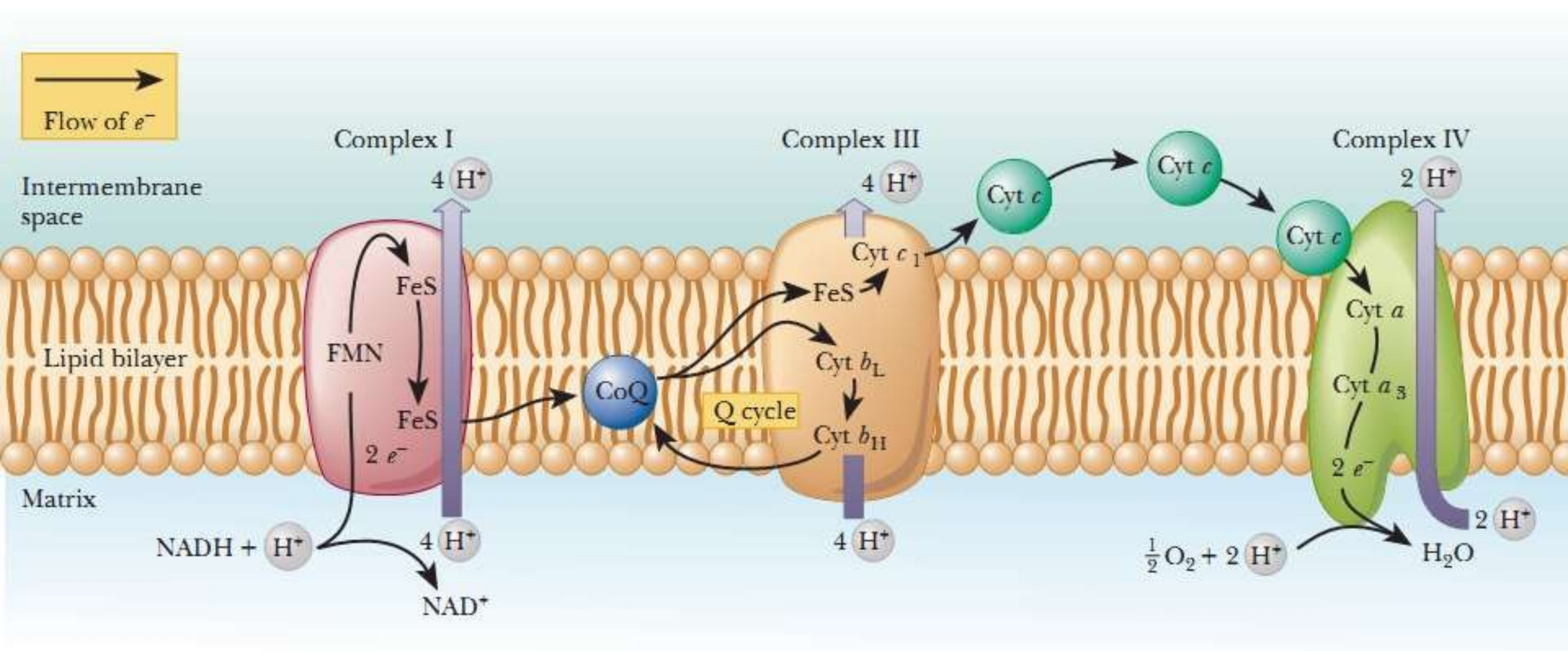
- **Aka Ubiquinol-cytochrome c oxidoreductase or cytochrome *bc_1* complex**
- **Consists of 3 main subunits: cyt. c1, cyt. b, and the Rieske Fe- S protein (ISP)**
- **Oxidises CoQ(QH2) in the membrane and reduces a mobile cyt. c on the exterior surface**
- **Transports $4H^+$ ions from the matrix to the cytosol**

Complex IV



- **Aka cytochrome c oxidase**
- **Consists of 13 subunits: 3 are encoded by the MtDNA and the remainder by the NuDNA**
- **Contains 2 heme groups, *heme a* and *heme a_3*, and 3 Cu ions (arranged as 2 copper centres Cu_A and Cu_B)**
- **Oxidizes reduced cytochrome c produced by complex III and transfers electrons from it to molecular O_2**
- **Translocate $2H^+$ ions across the mitochondrial membrane**

The Creation of a Proton Gradient During Electron Transport



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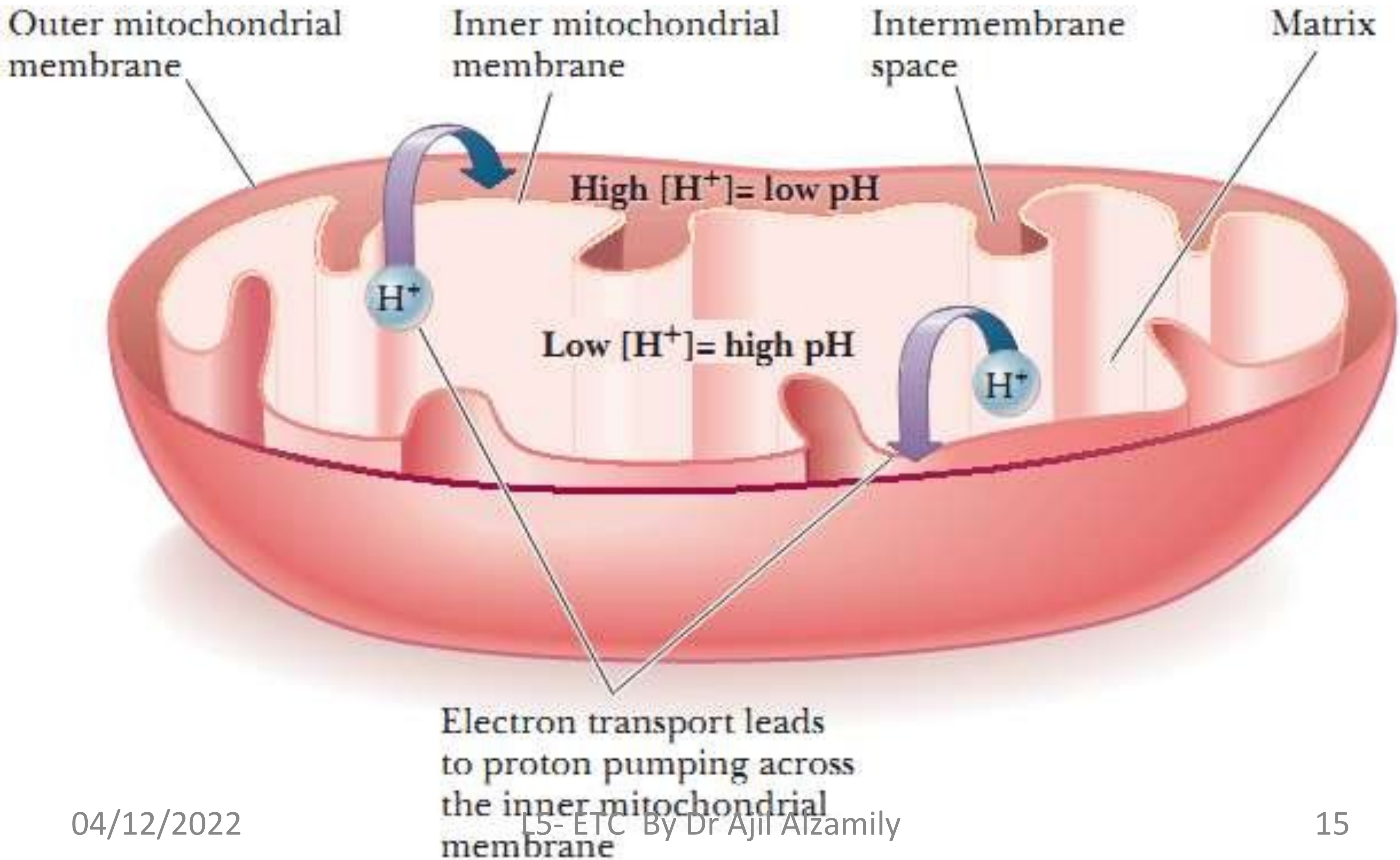
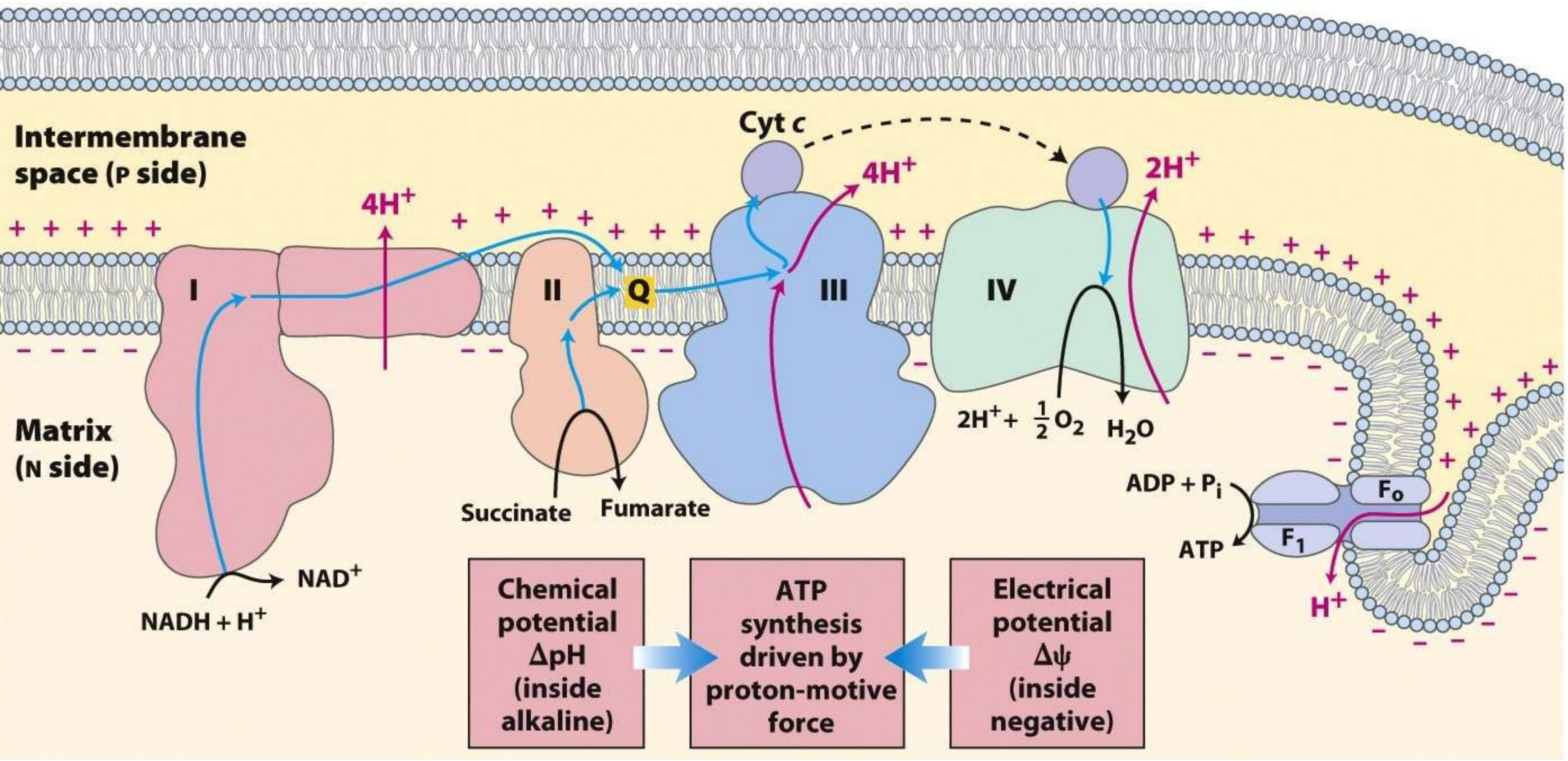
Proton Gradient as a result of electron Transport



Overview of Oxidative Phosphorylation



The Chemiosmotic Theory of Oxidative Phosphorylation

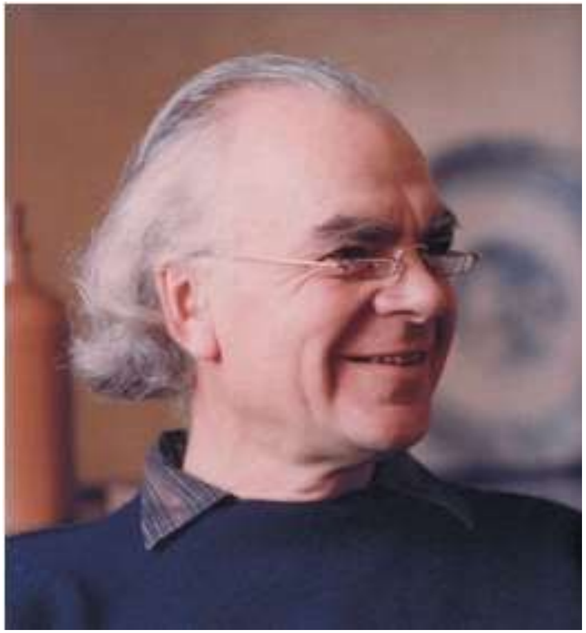
1st proposed in 1961 by a British Biochemist Peter Mitchell

- Widely accepted theory
- Based on the following postulates
-

– Electron transport through ETC generates H^+ gradient across the IMM

– H^+ gradient generates a proton motive force (PMF) which links ETC (Oxidative) and ATP synthesis (Phosphorylation)

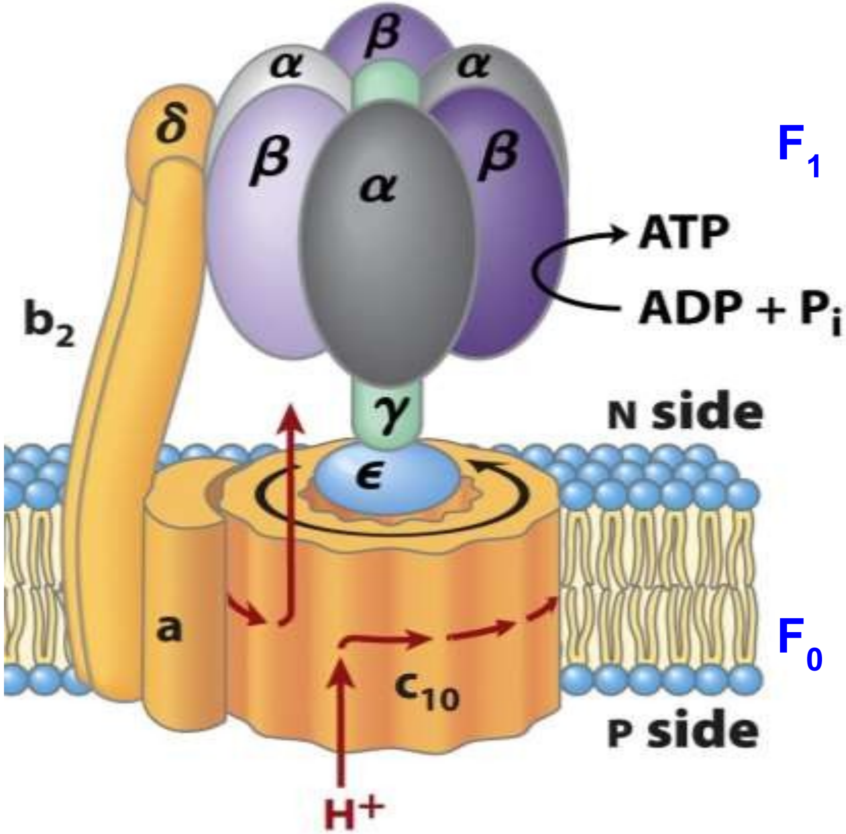
When H+ flow back to the matrix through ATP synthase to equalize the distribution, PMF drives the ATP synthesis



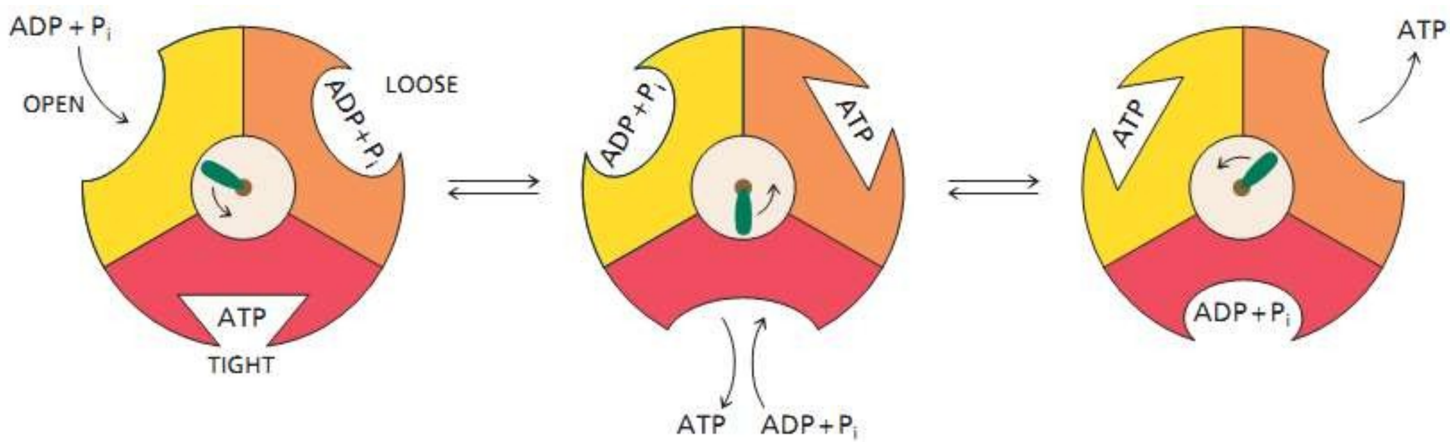
Peter Mitchell (1920– 1992)
Nobel Prize in 1978

ATP Synthase Is Composed of F_1 & F_0 Units

- An enzyme, a H pump,& a rotating molecular motor Synthesizes nearly all the cellular ATPs
- Consists of 2 units: F_1 and F_0
- F_1 consists α3, β3, γ, δ & ε subunits, andcatalyzes ATP synthesis
- F_0 unit is an aggregate of integral membrane proteins
- Consists of 3 subunits: a, b and c
- Forms a transmembrane pore through which H^+ ions move to drive ATP synthesis



Binding Change Mechanism for ATP Synthesis



Involves following three steps:

1. **1 mol of ADP and 1 mol of Pi bind to an O site**
2. **Rotation of γ shaft causes each of the 3 catalytic sites to change conformation**
   - **O site (with bound ADP & Pi) becomes a L site**
   - **L site (with ADP & Pi) becomes T site & form new ATP**
   - **T site containing ATP becomes an O site**
3. **ATP is released from the O site & ADP and P_i condense to form ATP in the T site.**

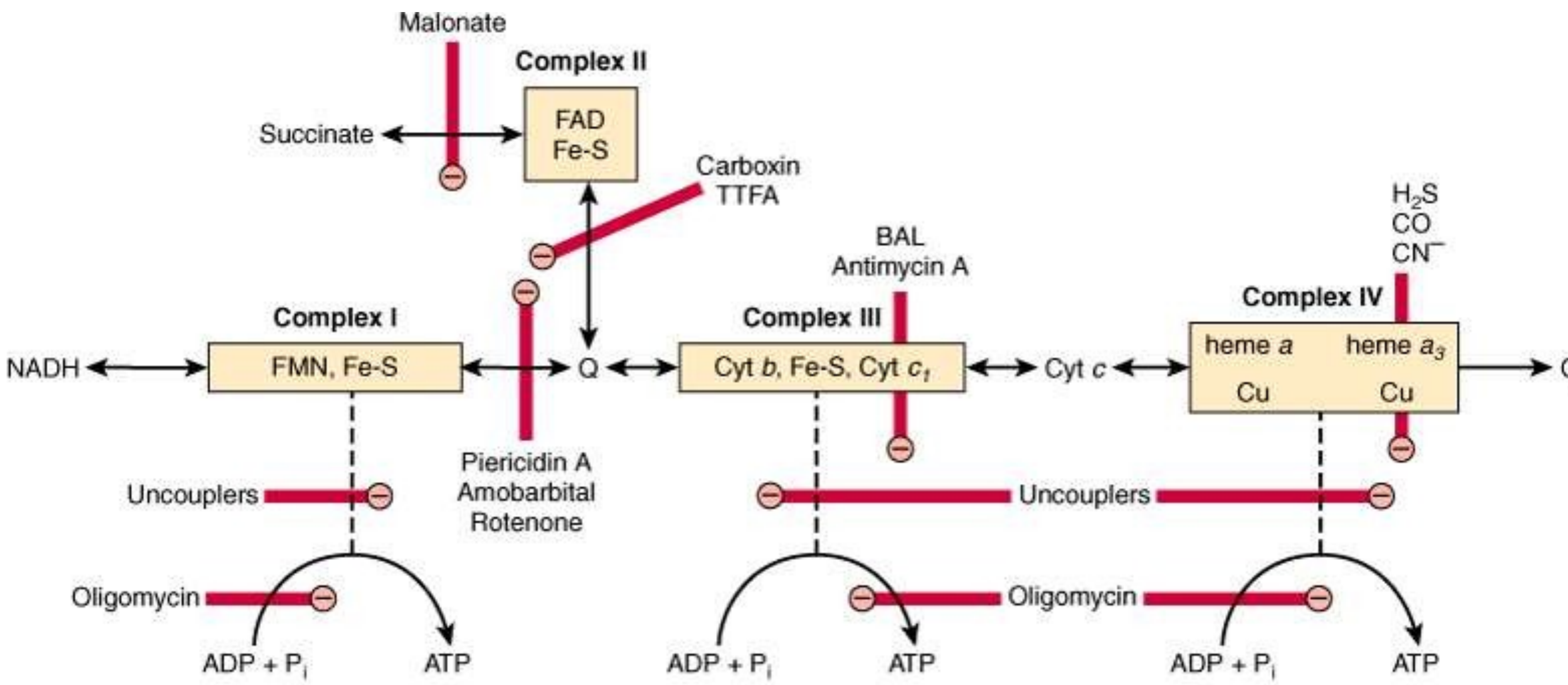
ATP Yield from Complete Oxidation of One Glucoses Molecule

Process	Direct Product	Final ATP	Final ATP (old concept)
Glycolysis	2NADH (cytosolic)	3 or 5*	4 or 6*
	2 ATP	2	2
Pruvate Oxidation (2/glucose)	2 NADH Mitochondrial Matrix	5	6
Acetyl CoA oxidation in TCA cycle (2/glucose)	6NADH (Mitochondrial Matrix)	15	18
	2FADH2	3	4
	2ATP or GTP	2	2
Total yields per glucose		30 or 32	36 or 38
*No. depends on which shuttle system transfers reducing equivalent in the mitochondria			

Inhibitors of ETC & Oxidative Phosphorylation

Types	Mode of action	Examples
Inhibitors of ETC	Block e^- transfer between different complexes & prosthetic groups of ETC	Amobarbitol, Rotenone, Piericidin A, Dimercaprol (BAL), Antimycin A, CN^- & CO
Uncouplers of e^- Transport from OP	Carry H^+ across the IMM, down their concentration gradient	2,4-DNP, Salicylate, Valinomycin,Gramicidin
Inhibition of ATP Synthase	Inhibit ATP synthase activity by blocking H^+ flow through F_0 channel	Oligomycin, Dicyclohexylcarbodiimide (DCCD)
Inhibition of ATP Export	Inhibits OP by inhibiting an enzyme adenine- nucleotide translocase	Attractylosides

Sites of Inhibitions of ETC and Oxidative Phosphorylation



Cyanide Poisoning

- CN^- is one of the most & rapidly acting poison known
- Individuals can die within a few minutes of exposure
- Acute inhalation of high concn. of CN^- can cause light headedness, breathlessness, dizziness, numbness & headaches
- CNS is the primary target of CN^- toxicity
- Cyanide is present in different forms e.g.

1. HCN in the air (fire smoke, cigarette smoke, automobile exhaust)
2. Cyanide salts (e.g. NaCN) in soil and water
3. Cyanoglycosides in foods (Almonds, stone fruits, sorghum, soyabean)

Biochemical Basis of CN^- Poisoning

CN^- binds to Iron in the heme of the cytochromeoxidase (IV)

Prevents electron transport to O_2 No

↓

mitochondrial respiration (ETC) No energy

↓

(ATP) production

↓

Rapid cell death

Since O_2 consumption is blocked, venous blood is as red as arterial blood

- What is phosphorylation?
- What is Oxidative phosphorylation?

- Formation of ATP from coupling of ADP and Pi is known as phosphorylation.
- Three types-
  - (1) Substrate level phosphorylation
  - (2) Photophosphorylation
  - (3) Oxidative phosphorylation

(1) Substrate level phosphorylation

- Enzymatic Transfer of phosphate from substrate to ADP to form ATP.
- ATP made in glycolysis and the TCA cycle is the result of substrate- level phosphorylation

(2) Photophosphorylation

- Formation of ATP in light reaction of photosynthesis.

- In which photosynthetic organisms capture the energy of sunlight—the ultimate source of energy in the biosphere and harness it to make ATP

- Photophosphorylation involves the *oxidation* of H_2O to O_2, with NADP as ultimate electron acceptor; it is absolutely dependent on the energy of light.

Oxidative phosphorylation

- How cells convert the stored metabolic energy of NADH and [FADH2] into ATP?

NADH or $FADH_2$-dependent ATP synthesis is the result of **oxidative phosphorylation.i. e. Formation of ATP from the oxidation of** NADH or $FADH_2$ in presence of oxygen through electron transport chain is know as **oxidative phosphorylation.**

Oxidative phosphorylation is the collection of energy yielding metabolism in aerobic organisms.

All oxidative steps in the degradation of carbohydrates, fats, and amino go through this final stage of cellular respiration, in which the energy of oxidation drives the synthesis of ATP

- Electrons stored in the form of the reduced coenzymes, NADH or [FADH2], are passed through an elaborate and highly organized chain of proteins and coenzymes, therefore called **electron transport chain,** finally reaching O_2 (molecular oxygen) is the terminal electron acceptor.

- Each component of the chain can exist in (at least) two oxidation states, and each component is successively reduced and reoxidized as electrons move through the chain from NADH (or [FADH2]) to O_2.

- In the course of electron transport, a proton gradient is established across the inner mitochondrial membrane. It is the energy of this proton gradient that drives ATP synthesis.

- In eukaryotes, oxidative phosphorylation occurs in inner mitochondrial membrane, and in prokaryote occurs in plasma membrane.

- Oxidative phosphorylation involves the *reduction* of O_2 to H_2O with electrons donated by NADH and $FADH_2$.

- It occurs equally well in light or darkness.

Thank you for your attention