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The Oxidative Phosphorylation in the course of Study Related to the Biological Science at Various Level

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Abstract **- Oxidative phosphorylation is the metabolic pathway in which cells use enzymes to oxidize nutrients thereby releasing energy in order to produce adenosine triphosphates (ATP) in eukaryotes; this takes place inside the mitochondria. Almost call aerobic organisms carry out oxidative phosphorylation. The energy stored in the chemical bonds of glucose is released by the cell in the citric acid cycle producing carbon dioxide, and the energetic electro donors NADH and FADH. Oxidative phosphorylation uses these molecules and oxygen (O_2) to produce ATP which is used throughout the cell whenever energy is needed. During oxidative phosphorylation, electrons are transferred from the electron donors to a series of electron acceptors in a series of redox reactions ending in oxygen, whose reactions releases half of the total of energy in eukaryotes have redox reactions are catalyzed by a series of complexes within the inner membranes of the cell's mitochondria.**

Keywords: Oxidation, Reduction, Phosphorylation, Mechanism, P/o Ratio.

I. Introduction

The endergonic synthesis of ATP from ADP and Pi in mitochondria, which is catalyzed by proton translocating ATP synthesis, is driven by the electron transport process. Yet, since complex of electron transporting reaction is physically distinct from the proteins mediating electron transport, the free energy released by election transport must be conserved in a form that ATP synthase can utilize. Such energy conservation is referred to as energy coupling or energy transduction.

The physical characterization of energy coupling proved to be surprisingly elusive; many sensible and often ingenious ideas have failed to with stand the test of experimental scrutiny. In this section it is examined some of the hypotheses that have been formulated to explain the coupling of electron transport and ATP Synthesis. Then it is explored that the coupling mechanism that has garnered the most experimental support, analyze the mechanism by which ATP is synthesized by ATP synthase.

II. Mechanisms of Oxidative Phosphorylation

The enzymes concerned in electron transport and oxidative phosphorylation are very complex and they are embedded in the inner mitochondrial membrane, rendering the detailed study of their interactions difficult. However, 3 principal hypotheses have been advanced to account for the coupling of the oxidation and phosphorylation. In other words, these hypotheses explain how the energy transfer between electron transport and ATP synthesis takes place.

1. Chemical coupling Hypothesis

This hypothesis postulates direct chemical coupling at all stages of the process. It is similar to the concept in glycolysis which states that the ATP produced in oxidative phosphorylation results from an energy-rich intermediate encountered in electron transport. Specifically, when an oxido-reduction reaction occurs between Ared and Boxi, the factor I is incorporated into the formation of an energy - rich structure Axi-I where indicates a linkage having an energy rich nature.

$$A_{red} + I + B_{oxi} \Leftrightarrow A_{oxi}\sim I + B_{red}$$

In subsequent reactions, an enzyme (E) replaces A_{oxi} in the compound $A_{oxi}\sim I$ to form an energy rich E~I. Later, inorganic phosphate reacts with E~I to form phosphoenzymes complex E~P containing the energy - rich enzyme-phosphate bond:

$$A_{oxi}\sim I + E \Leftrightarrow A_{oxi} + E\sim I$$

$$E\sim I + Pi \Leftrightarrow E\sim P + I$$

The enzyme - phosphate component finally reacts with ADP to form ATP.

$$E\sim P + ADP \Leftrightarrow E + ATP$$

Oxidative phosphorylation occurs in certain reactions of glycolysis, in the citric acid cycle and in the respiratory chain. However, it is only in those phosphorylations occurring at the substrate level in glycolysis and the citric acid cycle that the chemical mechanisms involved are known. Followings are three such equations is

3-phosphoglyceraldehyde + NAD^+ + Pi $\Rightarrow$ 1~3-biphosphogycerate + NADH + H^+

1~3-biphosphogycerate +ADP $\Rightarrow$ 3-phosphoglycerate +ATP

2-phosphoglycerate $\Rightarrow$ 2~phosphoenolpyruvate

2~phosphoenolpyruvate + ADP $\Rightarrow$ Pyruvate + ATP

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α-ketoglutarate + NAD^+ + CoA ⇒ Succinyl~CoA + NADH + H^+

Succinyl~CoA + GDP + Pi ⇒ Succinate + GTP

2. Conformational Coupling Hypothesis

In mitochondria, those are actively phosphorylating in the presence of a excess of ADP, the inner membrane pulls away from the outer membrane and assumes a condensed state". In the absence of ADP, the mitochondria have the normal structure or the "swollen state", in which the cristae project into the large matrix. The propounders of this hypothesis believe energy released in the transport of electrons along the respiratory chain causes the conformational changes.

3. Chemiosmotic Coupling Hypothesis

Peter Mitchell (1961) proposed that electron transport and ATP synthesis are coupled by a proton gradient, rather than by a covalent high-energy intermediates or an activated protein. According to this model, the transfer of electrons through the respiratory chain results in the pumping of protons (H+) from the matrix side (M-sides to the cytosol or cytoplasmic side (c-side) of inner mitochondrial membrane. The concentration of H^+ becomes higher on the cytoplasmic side, thus creating an electrochemical potential difference. This is positive on the cytoplasmic side. It is further proposes that the H^+ ions, ejected by electron transport, of low back into the matrix through a specific H+ channel or 'pore' in the FoF, ATPase molecule, driven by the concentration gradient of H^+. The free energy released, as proton (H^+) flows back through the ATPase, causes the coupled synthesis of ATP from ADP and phosphate by ATP synthetase. The inner mitochondrial membrane must be intact in the form of a completely closed vesicle, since an H^+ gradient across the inner membrane could noth otherwise exist. If however, a 'leak' of proton across the membrane is induced by uncouplers, the proton gradient would be discharged and consequently energy - coupling would file.

According the chemiosmotic hypothesis, the high energy chemical intermediates are replaced by a link between chemical processes: and the transport process – hence the phrase, chemiosmotic coupling. As the high-energy electrons from the hydrogen's of NADH and $FADH_2$ are transported down the respiratory chain in the mitochondrial inner membrane, the energy released as they pass from one carries molecule to the next is used to pump protons (H^+) across the inner membrane from the mitochondrial matrix into the inner membrane space. This creates an electrochemical proton gradient across the mitochondrial inner membrane, and the back flow of ret down this gradient is, in turn used to drive the membrane- bound enzyme ATP syntheses, which catalyzes the conversion of oxidative phosporylation.

III. The P/o Ratio: Energetic of Oxidative Phosphorylation

It was observed that for a pair of electrons entering the respiratory chain as NADH and traversing the entire chain to O_2 the standard free energy change, ΔG^o, is -220 kj/mol.

$$NADH + H^+ + ½O_2 \Leftrightarrow NAD^+ + H_2O$$

$$\Delta G^{01} = -nF\Delta E^0 = -2\ (96485)\ (-0.82-(-0.32)) = -220\ kJ/mol$$

The P/o ratio is the number of molecules of ATP synthesized per pair of electrons carried through electron transport ATP synthesis is quantitated as phosphate incorporation into ATP, and electron pairs are quantitated as oxygen uptake, in μ mol of o atoms reduced to water.

It may be imagined, that the precise measurements of oxygen consumption and ATP synthesis in a mitochondrial preparation are difficult to obtain, and many experimental pit falls can lead to inaccurate estimates of the p/o ratio. Early experiments suggested that the mitochondrial oxidation of NADH proceeds with a P/o ratio of 3 and oxidation of succinct proceeds with a P/o ratio of 2. However, as researchers got better at the preparing intact mitochondria and measuring oxygen consumption and ATP synthesis, it become clear that the P/o ratios were not integers. The general consensus now is that the P/o ratio is ~2.5 for oxidation of NADH and ~1.5 for oxidation of succinate. Indeed, these non integer P/o values contributed to the realization that phosphorylation and oxidation are not directly coupled. This mechanism of indirect coupling between oxidation and phosphorylation does not require an integral stoichiometric relationship between equivalents consumed and ATP synthesized.

$$NADH + H^+ + ½O_2 + 2.5ADP + 2.5P_1 \Leftrightarrow NAD^+ + H_2O + 2.5ATP$$

ΔG for ATP hydrolysis under intracellular conditions is estimated to be -50 kJ/mol or more, so the synthesis of 2.5mol ATP requires at least 2.5 x 50 kJ =125 kJ.

IV. Thermodynamic Implications of Chemiosmotic Coupling

According to Mitchell's hypothesis the energy coupling drives ATP synthesis by means of an electrochemical gradient, The energy is stored in this gradient, for the trans membrane flow of protons across the inner membrane, it may be written as follow:-

$$H^+_{in} \Rightarrow H^+_{out}$$

The free energy difference for protons across the inner mitochondrial membrane includes a term for the concentration difference and a term for the concentration difference and a term for the electrical potential. This is expressed as

$$\Delta G = RT\ ln\frac{[C_2]}{[C_1]} + zf\Delta\varphi$$

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C_1 & C_2= Proton concentrations on the two sides of the membrane,

Z = charge on the proton.

f = Faraday's constant

$\Delta\varphi$ = Potential difference across the membrane.

$$\Delta G = RT\ ln\frac{[H_{in}^{+}]}{[H_{otu}^{+}]} + zf\Delta\varphi$$

In terms of the matrix and cytoplasm pH values, the free energy differences is

$$\Delta G = -2.303\ RT\ (pH_{out} - pH_{in}) + f\Delta\varphi$$

Reported values for $\Delta\varphi$ and ΔpH vary, but the membrane potential is always found to be positive outside and negative inside and the pH is always more acidic outside and more basic inside. Taking typical values of $\Delta\varphi$ =0.18V and ΔpH = 1 unit, the free energy change associated with the movement of one mole of protons from inside to outside is

$$\Delta G = 2.3RT + f(0.18V)$$

With f = 96.485 kJ/V + mol, the value of ΔG at 37°C

$$\Delta G = 5.9\ kJ + 17.4\ kJ = 23.3\ kJ$$

Which is the free energy change for movement of a mole of protons across an inner membrane? The free energy terms for both the pH difference and the potential difference are unfavourable for the outward transport of protons, with the latter term making the greater contribution on the other hand; the A G for inward flow of protons is -23.3 kJ/mol. It is this energy that drives the synthesis of ATP, in. accord with Mitchell's model.

V. Discussion, Conclusion and Recommendations

The mechanism by which the free energy released by the electron-transport chain is stored and utilized in ATP synthesis is described by the chemiosmotic hypothesis. This hypothesis states that the free energy released by electron transport is converted by the generation of an electrochemical proton gradient across the inner mitochondrial membrane, which is harnessed to synthesize ATP. The proton gradient is created and maintained by the obligatory out war translocation of it across the inner mitochondrial membrane as electrons travel through following complexes of electron transport.

(i) Complex I (NADH - CoQ REDUCTASE)
(ii) Complex III (CoQ - CYTOCHROME c REDUCTASE),
(iii) Complex IV (CYTOCHROME C OXIDASE).

Complex III pumps protons via a redox loop mechanism called the 8 cycle, a bifurcated double cycle in which one molecule of CoQ H_2 is oxidized to CoQ H_2 is oxidized to CoQ and then is rereduced to CoQ H_2 by a second molecule of $CoQH_2$ in a process that collectively transfers tour on the inside to the outside while oxidizing one molecule of CoQ H_2 to CoQ. Electrons are transferred between the two CoQ's which are bound at Qo and cytochrome e, via the ISP, which under goes a conformational change in doing so. Complex IV contains no (H^++e^-) carries such as $CoQH_2$ and hence translocates proteins via a proton pump mechanism. Bacteriorhopodism, the best characterized proton pump, translocates protons with driven process. This involves a trans to cis isomerization of bacteriorhopsin's retinal prosthetic group on absorbing a photon, followed by the translocation of a proton through the hydrophilic central channel of this trans membrane protein via a process that involves conformational and pK changes of the polar groups lining the channel as the retinal relaxes to its ground state. Complex is through to pump protons via similar mechanism that is driven by the changes in the redox state of its heme a_3-Cu_B binuclear centre as it reduces O_2, to H_2O.

The energy stored in the electrochemical proton gradient is utilized by proton translocating ATP synthase (complex V, f_i f_o- ATPase) in the synthesis of ATP via the binding change mechanism, by coupling this process to the exergonic transport of H^+ hock to the inside. Mitochondrial proton - translocating ATP synthase consists of two oligomeric components : $F_1(\alpha,\beta,\gamma\delta E)$, a peripheral membrane protein that appears as "lollipops" in electron micrograph*s* of the inner mitochondrial membrane, and F_o, an integral membrane protein that contains the proton channel. The conformational changes that promote the synthesis of ATP from ADP + Pi arise through the demonstrated rotation of the γ subunit relative to the catalytic $\alpha_3\beta_3$ assembly that contains the enzyme's three of active sites. The γ subunit is attached to a ring of c subunits in F_o whose rotation is driven by the passage of protons between it and the α subunit.

Most of the energy captured for ATP synthesis from oxidative reactions cells is generated in mitochondrial oxidative phosphorylation. Reduced electron carriers, both NADH and $FADH_2$, shuttle their reducing equivalents into the mitochondrial matrix. Enzyme complexes bound to the inner mitochondrial membrane pass these electrons through the respiratory chain, a series of electron carriers of ever-increasing reduction potential. The complexes are numbered I (NADH dehydrogenase) II (Succinate-coenzyme Q oxidoreductase), III (Coenzyme Q-Cytochrome c oxidoreductase), IV (cytochrome oxidase) and V (ATP synthase). Electrons are eventually transferred to O_2 which is reduced to water. The reactions of complexes I, III and IV provide energy to pump protons through the inner membrane making the outer surface much more acidic than the matrix. Discharge of the resultant proton gradient, when back into the matrix through a specific ion channel generates energy that is used to drive ATP synthesis. Although respiration accounts for about 90% of the total oxygen uptake in

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most cells, in numerous other reacting dozens of enzymes use O_2 as a substrates-oxygenases, oxidases, and hydroxylases. Same reactions generate partially reduced oxygen species-hydroxyl radical super oxide, and peroxide - which are toxic and mutagenic. Cells possess numerous mechanisms for detoxification of these reactive oxygen species.

The P/o ratio is the number of molecules of ATP formed in oxidative phosporylation per two electrons flowing through a defined segment of the electron transport chain. The consenses value for the mitochondrial p/o ratio is 10/4, or 2.5 for electrons entering the electron transport chain as NADH. For succinate to O_2, the P/o ratio in this case would be 6/4 or 1.5.

It is concluded that phosphorylation occurs as electrons make their way along the electron-transport chain. The phosphorylation occurs when reduced cytochrome c is oxidized by molecular oxygen by the enzymes in complex IV, Similarly, complex I and III, but not complex II contain phosphorylation sites and, ADP and H_3PO_4 cure converted to ATP as electrons move along the carriers in those complexes. In complex, there is at least one carrier whose E_o' depends on the concentration of ATP. These carriers [$(Fe\text{-}S)_4$ in complex I; b_{562} in complex III; a_3 in complex IV] have therefore been proposed as the carrier specifically associated with the phosphorylation site in the complex.

Since the life Sciences along with the physical sciences have received much more attention to revise and refresh course there is an urgent need now to lay emphasis on the protection of environment all over the world and to provide a better life to the living beings of this planet in order to maintain a environment and proper balance between the environment and human survival it becomes imperative to incorporate concepts of immediate concern that have direct complications not only to theory but practical work and their subsequent application for environmental protection and human survival maintain the ecological and biochemical to maintain and then the ecological balance.

The study of such concept as oxidative phosphorylation should be specifically introduced the course of study related to the biological Sciences at various levels.

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