BASIC SCIENCE ASPECTS OF THE MITOCHONDRIA
SECTION I

OVERVIEW OF MITOCHONDRIAL STRUCTURE AND FUNCTION

Mitochondria are organelles found in virtually all eukaryotic cells, animal and vegetable, where they perform similar, if not absolutely identical, functions in the cell and the organism.

They are believed by many scientists to be independent living organisms, which live in symbiosis with the cells they occupy. They have their own genome and reproduce independently of the cells' reproductive cycle. The functions they perform will be discussed below, after the structure of the organelle is discussed. While their size and shape may vary greatly, all mitochondria are organelles consisting of two membranes enclosing two spaces.

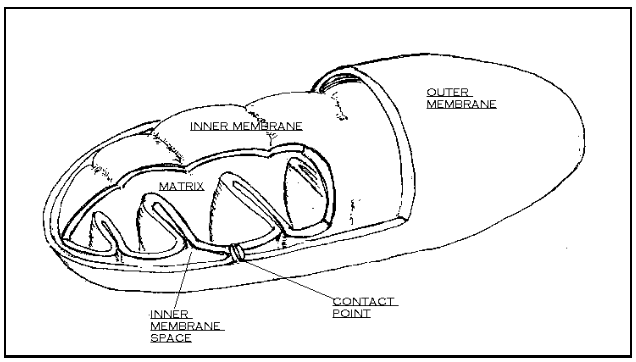

Figure 1

The outer membrane is a bi-lipid membrane quite similar to other cellular membranes. Contained within their outer membrane is the enfolded inner membrane which is quite different from the outer membrane and other cellular membranes. This inner membrane and the enzymes embedded or floating in it, surround the matrix which is a dense liquid containing many enzymes and the genome of the organelle.

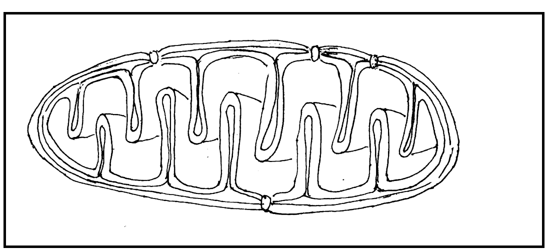
Figure 2a

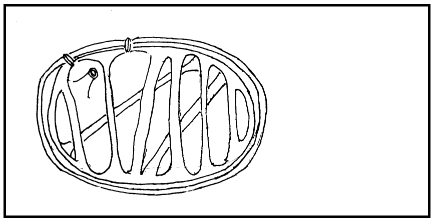

Figure 3a

The outer membrane is quite permeable, whereas the inner membrane is largely impermeable; three quarters of its volume is made up of proteins. It is here and in the matrix that the work of the mitochondria is carried out.

The inter membrane space between the outer and inner membranes is generally about like the cytosol due to the permeability of the outer membrane, and the two membranes are joined at several anchor pores called contact points which anchors the inner membrane. These points are also pores for the entry of nuclear encoded proteins and pre-proteins into the matrix of the organelle.

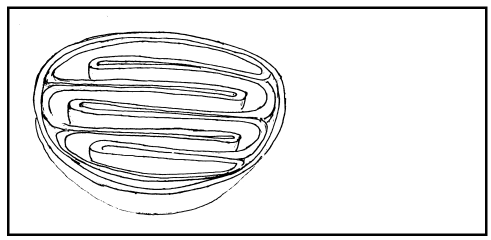

Figure 3b

The infolding of the inner membrane forms the cristae, which assume varied shapes, the original descriptions of these were of the classic baffle type. More advanced types of microscopic imaging have shown that in some mitochondria, the cristae are

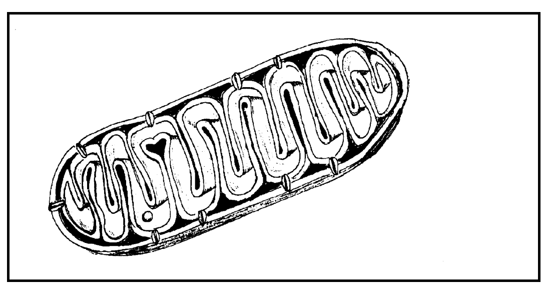

Figure 2b

tubular and in some cases the in-foldings are longitudinal rather than lateral.

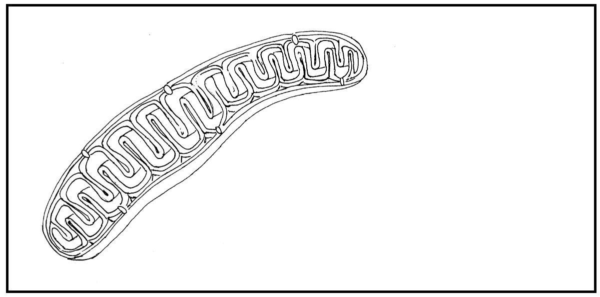
Figure 3d

None of these differences have much effect on their functions which will be discussed below.

In general, mitochondrial structure is quite similar to that of a bacteria, leading most scientists to the belief that mitochondria are bacteria which were engulfed or invaded eukaryotic cells in the past and established a mutually beneficial symbiotic relationship with these cells.

Mitochondria possesses a genome, several copies of which are contained in the matrix, and this genome is relatively small. Many scientists believe that at some time in the past, around 80% of the genes of the original bacterial genome were transferred to the nucleus of the cells, and only around 20% remains in the organelle. Many of the proteins used by the mitochondria are now encoded in the nuclear DNA and are transferred to the mitochondria from there through the pores at the contact points, after which they are incorporated into the structure. Most mitochondrial structure is a combination of a few proteins encoded in the mitochondrial genome and several encoded in the nuclear genome.

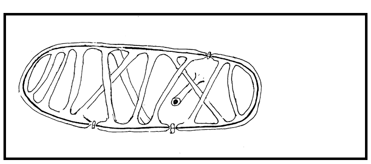
Figure 3c

Those encoded in the mitochondrial genome are maternally inherited, while those in the nuclear genome are subject to mendelian inheritance. This will be discussed in detail in a separate chapter.

Mitochondria reproduce by fission, and apparently are able to fuse and separate freely.

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GENERAL FUNCTIONS

Mitochondria have several important functions in the cell, the most well known being that of the production of Adenosine Triphosphate by the process of oxidative phosphorylation. Around 95% of the ATP produced in the body is produced in the mitochondria. For this reason, they are often referred to as the power house of the cell, although they have several other equally important functions as well.

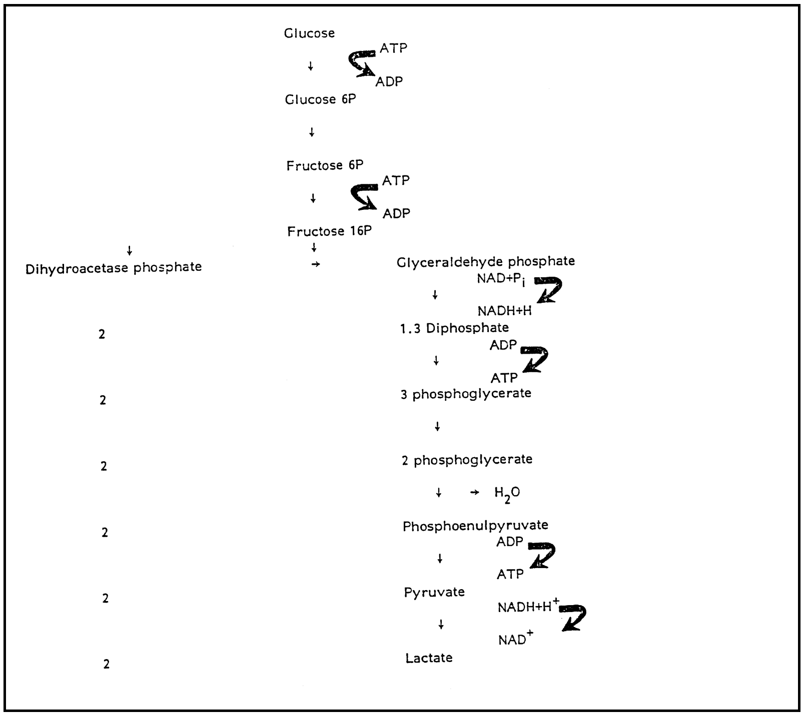

Figure 4

All molecules contain energy, stored in the molecular structure itself. A portion of that energy can be used to do work. This is called free energy. Oxidation of a molecule results in the release of free energy. Complete combustion (burning) of organic molecules, for example, releases all of the available free energy as heat. Reduction of a molecule requires an input of energy.

Energy can be transferred from one molecule to another by enzymes. The molecules that are converted by enzymes, that is, the reactants, are called substrates.

Nutrients are organic molecules that are ultimately derived from food sources. They start off as fats, carbohydrates, and proteins. Enzymes involved in intermediate metabolism oxidize nutrient molecules to a form that can be converted to energy by mitochondria. Fats, carbohydrates and proteins are broken down to individual fatty acids, simple sugars and amino acids.

Cells utilize glucose, and its breakdown products, as well as fatty and amino acids as fuel. These are broken down in the process of digestion and transported to the cells via the circulatory system where they diffuse from the capillaries through the intracellular space and into the cytosol where the first step, that of anaerobic glycolysis occurs.

This is the method by which some anaerobic organisms produce ATP for their energy needs.

The end product of anaerobic glycolysis is pyruvate, which is broken down to two carriers, Nicotinamide Adenine Dinucleotide (NAD) and Flavin Adenine Dinucleotide (FAD), which must be transported across the virtually impenetrable mitochondrial inner membrane to the matrix.

This is accomplished by two shuttle systems, the malate aspartate and glycero-phosphate shuttles, which deliver these co-enzymes to the matrix in the oxidized state.

In the matrix, these are processed in the Krebs or Citric Acid Cycle where they are reduced, that is to say, where they have additional hydrogen ions attached to them, and after this reduction, they are carried to the inner membrane for further processing by the electron transport chain, which is made up of five complexes and two carriers.

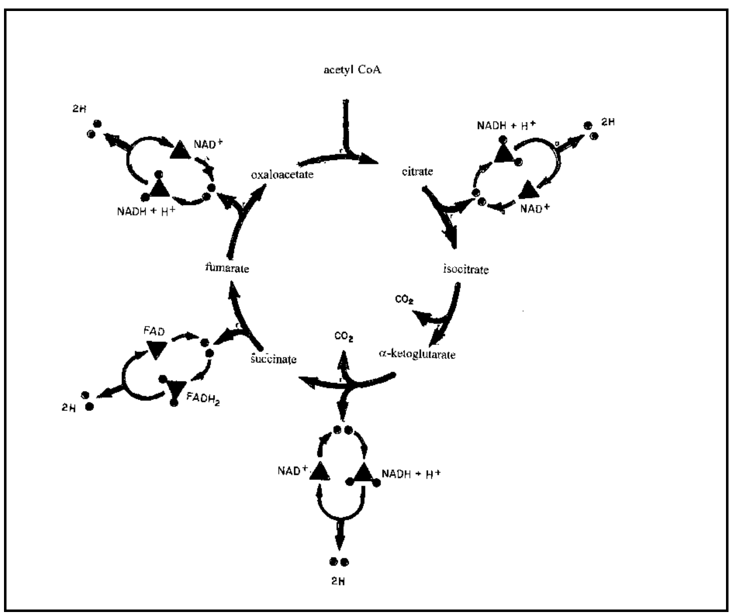

Figure 5

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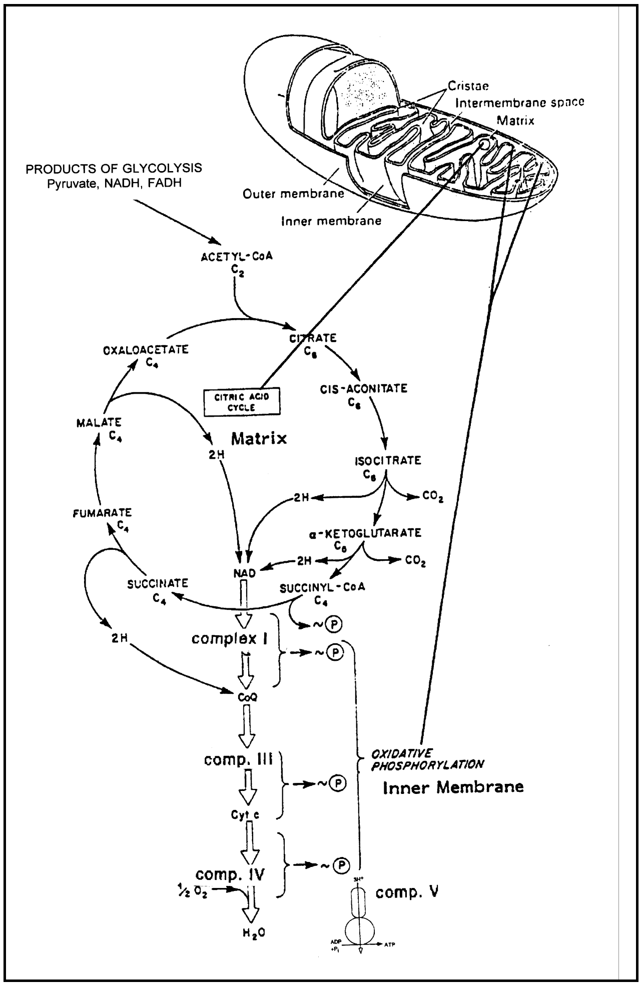

Figure 6

To understand this process, we should know the basic structure of the inner membrane, a bi-lipid membrane in which are embedded the complexes which carry out the electron transport function.

INNER MEMBRANE

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The inner membrane is much larger and more extensive than the outer membrane and is enfolded within the inner membrane, the enfoldings forming a cristae. The inner membrane in its mass consists of approximately 3/4 proteins, which are various transport mechanisms and the five complexes of the electron transport chain and while these are said to be embedded in the inner membrane, they actually float in the inner membrane. Textbook illustrations tend to show them in a straight line so one can understand how electrons are transported from one to the other; actually, they are not embedded in an orderly linear fashion, and are rather scattered. (Figures 8 & 9). Also embedded in the inner membrane are enzyme carriers such as co-enzyme Q10 and cytochrome c which accept electrons from one complex and transport them to another. These enzymes and complexes are extremely numerous and are in very close proximity to each other. Figures 7, 8 and 9 show a fairly accurate depictation of their

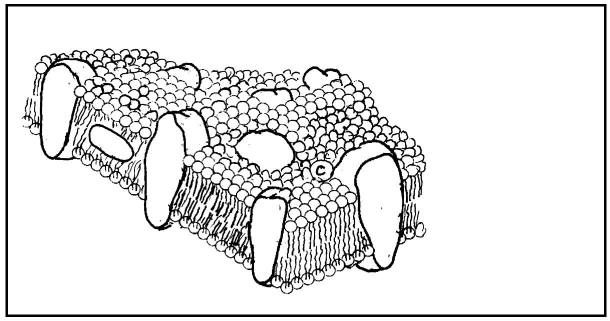

Figure 7

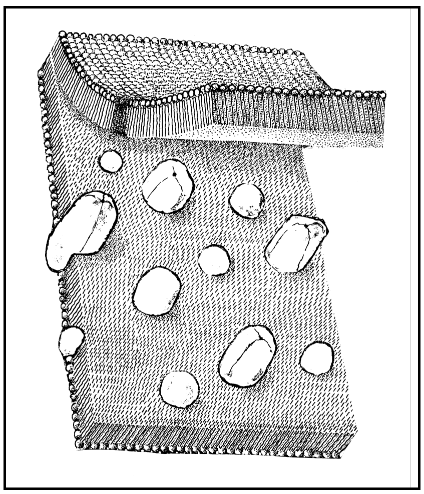

Figure 8

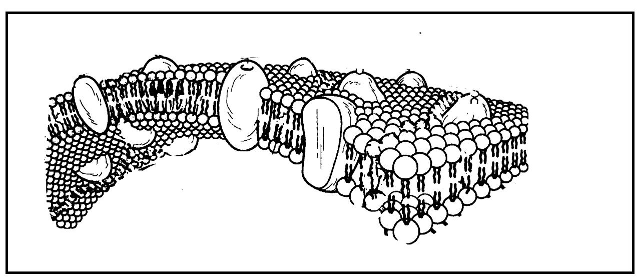

Figure 9

number and proximity to each other. Figure 10 is a more stylistic depictation showing the complexes and the various transport mechanisms involving moving substances in and out of the otherwise impermeable inner membrane. The inner membrane is permeable to gasses but without an import mechanism is impermeable to most other molecules. For adenosine diphosphate to be processed in oxidative phosphorylation, much of it must be moved across the inner membrane after

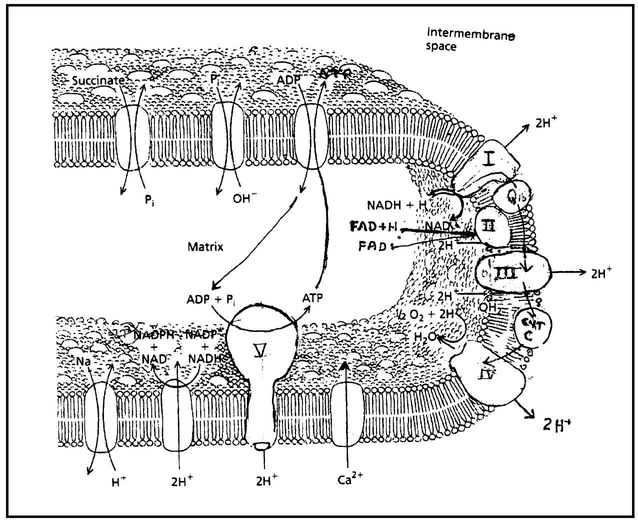

Figure 10

Adenosine Triphosphate is created it must be moved back out across the membrane to be used in other parts of the cell. The electron carrier enzymes NAD and FAD as mentioned above must be transported into the matrix by shuttles. (Figure 10)

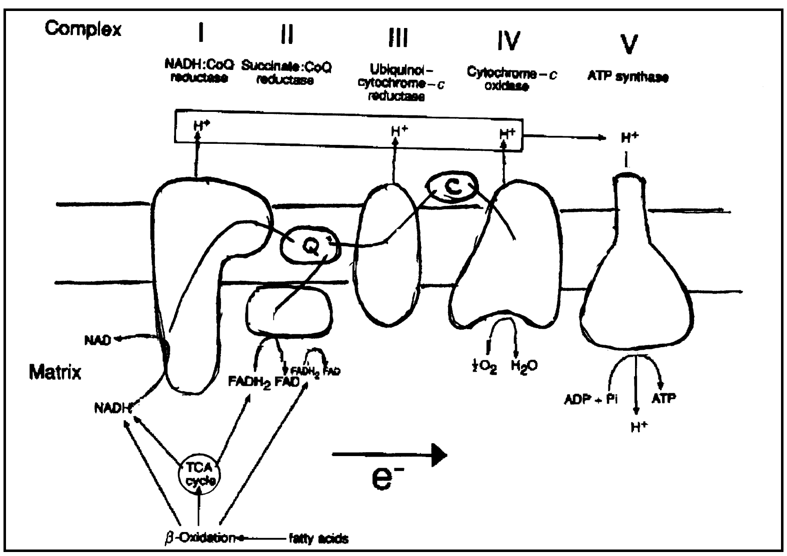

Figure 11

Complex I accepts fuel from the citric acid cycle in the form of NADH, which donates electrons to the chain. Part of the energy of these electrons are used

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to pump a proton across the inner membrane, after which the electrons are passed to Complex III via coenzyme Q. Complex II accepts electrons from FADH2 and also passes them to Complex III via coenzyme Q. Complex III uses another part of the energy of these electrons to pump another proton across the inner mitochondrial membrane. The electrons are then passed to Complex IV via cytochrome C where the remaining energy is used to pump the third proton across the membrane. The de-energized electron is then transferred to oxygen to generate water.

The relative excess of protons in the intermembrane space creates a pH and redox gradient across the inner mitochondrial membrane. The energy of this gradient, which is known as the proton motive force, is used by Complex V to convert adenosine diphosphate (ADP) to adenosine triphosphate (ATP), the chemical energy "currency" of the cell. ATP can then be transported to where work needs to be done. In this fashion, electron transport to oxygen in Complex IV is said to be coupled to oxidative phosphorylation in Complex V. If for any reason the electron transport system is diverted or does not create sufficient proton motive force to operate Complex V, then the electron transport chain and oxidative phosphorylation are said to be

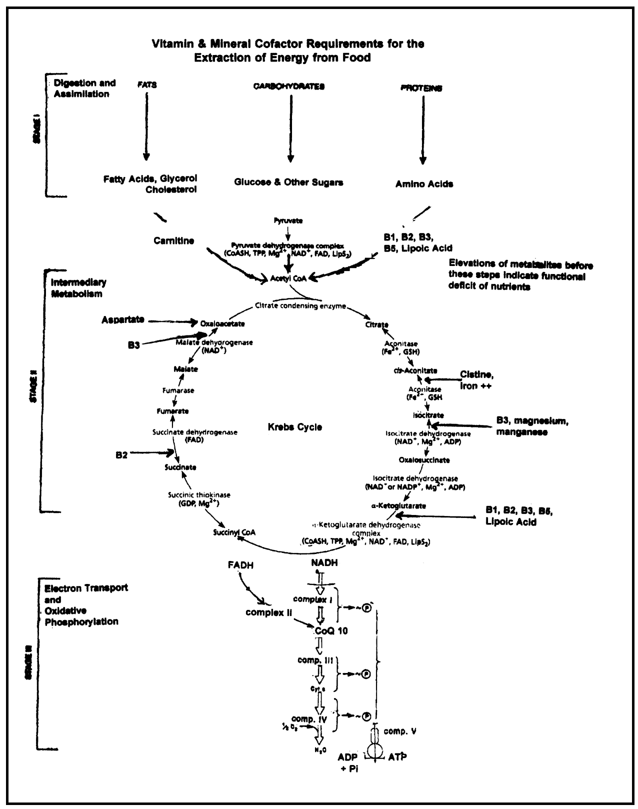

Figure 12

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uncoupled. The coupling and uncoupling of electron transport and oxidative phosphorylation will be discussed in a later chapter in detail. The coupling of electron transport to oxidative phosphorylation is essential for normal cell function and, ultimately, for the life of the cell.

Several nutrients and enzymes are involved in Krebs Cycle and electron transport chain functions. Deficiencies of these, may lead to dysfunctions. (Figure 12)

REFERENCES:

1. Alberts B, Bray D, Lewis J, Raff M, Roberts K, Watson JD. Molecular Biology of the Cell. 3rd ed. New York: Garland Publishing, 1994:1-1294.
2. Lodish L, Baltimore D, Berk A, Zipursky SL, Matsudaira P, Darnell J. Molecular Cell Biology. 3rd ed. New York: Scientific American Books, 1995:1-1344.
3. Gillham NW. Organelle genes and genomes. New York: Oxford University Press, 1994:424.
4. Cooper GM. The Cell, A Molecular Approach. Herndon, VA: ASM Press, 1997:650. 5. Hannavy K, Rospert S, Schatz G. Protein import into mitochondria: a paradigm for the translocation of polypeptides across membranes. Curr Opin Cell Biol 1993; 5:694-700.
6. Lithgow T, Click BS, Schatz G. The protein import receptor of mitochondria. Trends Biochem Sci 1995; 20:98-101.
7. Pfanner N, Craig EA, Meijer J. The import machinery of the inner mitochondrial membrane. Trends in Biochem Sci 1994; 19:368-372.
8. Glick B, Schatz G. Import of proteins into mitochondria. Annu Rev Genet 1991; 25:21-44.
9. Fawcett DW. An Atlas of Fine Structure: The Cell, Its Organelles and Inclusions. Philadelphia: W.B. Saunders, 1966:1-446.
10. Bereiter-Hahn J. Behaivior of mitochondria in the living cell. International Review of Cytology 1990; 122:1-63.
11. Ardail D, Lerme F, Louisot P. Involvement of contact sites in phosphatidylserine import into liver mitochondria. J Biol Chem 1991; 266:7978-7981.
12. Brdiczka D. Contact sites between mitochondrial envelope membranes. Structure and function in energy- and protein-transfer. Biochim Biophys Acta 1991; 1071:291-312.
13. Lea PJ, Hollenberg MJ. Mitochondrial structure revealed by high resolution scanning electron microscopy. Am J Anat. 1989; 184: 245-257.
14. Winslow JL, Hollenberg MJ, Lea PJ. Resolution limit of serial sections for 3D reconstruction of tubular cristae in rat liver mitochondria. J Electron Microsc Tech 1991; 18:241-248.
15. Mannella CA, Marko M, Buttle K. Reconsidering mitochondrial structure: new views of an old organelle. Trends in Biochem Sci 1997; 22:37-38.

16. Srere PA. The infrastructure of the mitochondrial matrix. Trends in Biochem Sci 1980; 5:120-121.
17. Robinson JB, Jr., Inman L, Sumegi B, Srere PA. Further characterization of the Krebs tricarboxylic acid cycle metabolon. J Biol Chem 1987; 262:1786-1790.
18. Barnes SJ, Weitzman PD. Organization of citric acid cycle enzymes into a multienzyme cluster. FEBS Lett 1986; 201:267-270.
19. Baker KP, Scaniel A, Vestweber D, Schatz C. A yeast mitochondrial outer membrane protein essential for protein import and cell viability. Nature 1990; 348:605-609.
20. Hatefi Y. Resolution of complex II and isolation of succinate dehydrogenase. Methods Enzymol 1978; LIII:27-35.
21. Hackenbrock CR. Lateral diffusion and electron transport in the mitochondrial inner membrane. Trends in Biochem Sci 1981; 6:151-154.
22. Klingenberg M, Winkler E, Huang S-C. ADP/ATP carrier and uncoupling protein. Methods Enzymol 1995; 260:369-389.
23. Zalman LS, Nikaido H, Kagawa Y. Mitochondrial outer membrane contains a protein producing nonspecific diffusion channels. J Biol Chem 1997; 255:1771-1774.
24. Brdiczka D. Function of the outer mitochondrial compartment in regulation of energy metabolism. Biochim Biophys Acta 1994; 1187:264-269.
25. Wicker U, Bucheler K, Gellerich FN, Wagner M, Kapischke M, Brdiczka D. Effect of macromolecules on the structure of the mitochondrial inter-membrane space and the regulation of hexokinase. Biochim Biophys Acta 1993; 1142:228-239.
26. Bucheler K, Adams V, Brdiczka D. Localization of the ATP/ADP translocator in the inner membrane and regulation of contact sites between mitochondrial envelope membranes by ADP. A study on freeze-fractured isolated liver mitochondria. Biochim Biophys Acta 1991; 1056:233-242.
27. Shiao YJ, Lupo C, Vance JE. Evidence that phosphatidylserine is imported into mitochondria via a mitochondria-associated membrane and that the majority of mitochondrial phosphatidylethanolamine is derived from decarboxylation of phosphatidylserine. J Biol Chem 1995; 270:11190-11198.
28. Hurt EC, Pesold-Hurt B, Schatz C. The amino-terminal region of an imported mitochondrial precursor polypeptide can direct cytoplasmic dihydrofolate reductase into the mitochondrial matrix. EMBO J 1984; 3:3149-3156.
29. Martin J, Mahlke K, Pfanner N. Role of an energized inner membrane in mitochondrial protein import. J Biol Chem 1991; 266:18051-58057.
30. Wienhues U, Neupert W. Protein translocation across mitochondrial membranes. Bioessays 1992; 14:17-23.
31. Ackerman SH, Gatti DL, Gellefors P, Douglas MC, Tzagoloff A. ATP13, a nuclear gene of Saccharomyces cerevisiae essential for the expression of subunit 9 of the mitochondrial ATPase. FEBS Lett 1991; 278:234-238.