PREFACE

While I can say that I was naïve about the task confronting me in starting the first edition of the book on mitochondria, I had no illusions about the challenge of following it with a second edition. Was there in fact a need for a second edition?

The greatest encouragement came from the reception of the first edition and from the many favorable and supportive comments from friends and fellow scientists over the past eight years. As projected, the book filled the need of a diverse group of researchers and students to gain a broad perspective on the subject and also served as an introduction to many intricacies of this broad field. It was intended to explain basic principles and insights that would not become obsolete, although many more details, supporting data, and specific examples could be anticipated to be published as the book was written and during the following years. While the first edition undoubtedly contained mistakes and omissions, there were no major errors and misinterpretations that kept me awake at night wishing to publish an immediate correction or retraction. Thus, the plan for a second edition could mature over several years, with careful considerations about what to modify and what to add.

From the outset it was clear that the focus had to be sharpened to keep the volume within bounds. Thus, most attention was paid to progress in the understanding of mammalian mitochondria. Information from the first edition about mitochondria in other organisms (fungi, protozoa, plants) was retained to illustrate the diversity of mitochondria and some of the unique aspects of mitochondria in these organisms. And, as pointed out before, pioneering studies in microorganisms such as yeasts have continued to provide important clues and insights that have guided the exploration of mitochondria in higher eukaryotes. Consequently, multiple references to such recent studies are included.

What is really new since the appearance of the first edition? First of all, literally hundreds of new, complete sequences of mtDNA from many different organisms have been added to the database, providing raw material for addressing questions on the evolution of this organelle over both long and shorter time scales. The relationship of hydrogenosomes and mitosomes to mitochondria has been clarified.

Many more details have been added to the understanding of the biogenesis of mitochondria, including the replication of mtDNA, the transcription of the mitochondrial genome, and pathways for protein import into various membranes and compartments of the organelle. An important new addition to this subject is the elucidation of the biogenesis of the iron–sulfur centers in mitochondria, one of the most essential functions contributed to the cell exclusively by this organelle.

The dynamic behavior of mitochondria, already apparent many years ago, is an active area in which much progress has been achieved with regard to components involved in fission and fusion, but much more needs to be learned, especially about the control of these processes. A functional connection between some of these proteins governing these normal morphological processes and their role in apoptosis is becoming apparent. Controversy or uncertainty continues in a detailed understanding of the two phenomena relating mitochondria to apoptosis: What is the mechanism of the release of cytochrome c and other proteins from the intermembrane space, and what is the nature of the pore/channel responsible for the permeability transition (breakdown of the (inner) membrane potential)?

High-resolution structures of some of the five complexes of the electron transport chain were included in the last edition of this book. The structure of complex II (succinate–ubiquinone oxidoreductase) from yeast and mammals can now be added to the list, and significant progress has been made with the recent publication of the structure of the peripheral subcomplex of the prokaryotic complex I. However, the role of ~31 "ancillary" subunits in the mammalian/eukaryotic complex continues to be a challenging puzzle. Significant progress has also been made in the elucidation of the complete structure of complex V. The structure of the c-ring has been solved for related enzymes, allowing more informed guesses about the translocation of protons and the mechanism inducing rotations. Some significant question marks remain. The existence of supercomplexes in the electron transport chain has been firmly established, challenging us to seek an understanding of their physiological significance.

An explosion of proteomic studies has emphasized the view that mitochon- re not just the powerhouse of the cell producing more or less ATP, demand, but their composition is highly variable from tissue to ecause of the many other functions contributed by mito- ialized cell types. Characterizing mitochondria from rat ts when one is interested in mitochondria from neurons. s been made in the identification and structural character-

ization of the large family of transporters, although the specificity of many family members remains to be established. The traffic of metabolites and ions firmly integrates mitochondria into many cellular activities. In this context the spotlight has in recent years been on potential regulatory mechanisms involving protein kinases, phosphatases, and their targets inside the mitochondria. Many intriguing observations suggest the presence of such mechanisms, but signaling pathways and specific targets remain to be identified and understood. For example, how (and why) does a defect in the mitochondrial PINK-1 kinase cause Parkinson's disease?

An avalanche of publications (>75,000 total, more than 3000 in 2006–2007 in PubMed) on reactive oxygen species (ROS) seems to lead to the paradox that "the more facts we learn, the less we understand the process we study." The reader can be referred to an illuminating essay by Y. Lazebnik (*Cancer Cell*, 2002) entitled "Can a biologist fix a radio?—Or, what I learned while studying apoptosis." The mitochondrial electron transport chain continues to be thought of as the major source of single electrons for superoxide formation, with complex II now added to complexes I and III as a source of the single electron. Then what happens? A low level of ROS may be a beneficial regulatory signal, but at higher levels it becomes a potentially pathological "oxidative stress" that can affect proteins, lipids, and DNA, whatever one's preference and particular focus. Experiments with animal models in which scavenger enzymes have been elevated or knocked out continue to emphasize the effects of ROS on life spans and well-being.

Finally, nitric oxide has emerged as a potential player in the control of mitochondrial activity. Its postulated functions range from being (1) an inhibitor of cytochrome oxidase, (2) a reaction with ROS to form peroxynitrite and nitrosothiols in proteins, and (3) a regulator of mitochondrial biogenesis via activation of guanylate cyclase.

Instead of "oxidative stress," one can also consider the "redox state" of a cell which could be reflected, for example, in NAD^+/NADH or GSH/GSSG ratios. Mitochondria are likely to be key players in regulating the relative levels of these small molecules; and in addition to modulating metabolic activity, they thus may influence chromatin remodeling and gene expression through NAD^+-dependent deacetylations of histones, for example. Such a mechanism provides a very direct cross-talk between mitochondria and the nucleus.

The number of "mitochondrial diseases" continues to expand, depending on the definition of the term. The original emphasis was on defects in oxidative phosphorylation due to mutations in mitochondrial DNA. Not surprisingly, nuclear mutations affecting subunits of the complexes of the electron transport chain must be included. However, where does one draw the line? In this book, some attention is given to (a) diseases resulting from defective mtDNA maintenance or replication and (b) Friedreich's ataxia, a problem in mitochondrial iron "metabolism." Defects in cardiolipin metabolism can also lead to a mitochondrial disease, because of the unique association of this lipid with mitochondrial membrane complexes. Finally, the involvement of mitochondria

in the development of neurodegenerative diseases such as Alzheimer's disease, Parkinson's disease, Huntington's disease, and amyotrophic lateral sclerosis (ALS) and in the general process of senescence (aging) remains an intriguing and attractive hypothesis, although a distinction must be made between a primary effect (due to specific mutations/polymorphisms in mtDNA) and secondary effects of aged mitochondria on other cellular housekeeping mechanisms such as proteasome activity (protein quality control).

There are great expectations for progress from a new (?) approach referred to as systems biology. Even without the hype, it is clear that a higher level of understanding biological phenomena must be derived not from understanding a single enzyme, or even a single pathway, but from an understanding of a larger ensemble of proteins and their activities. Mitochondria represent such an ensemble of proteins, contained within a well-defined compartment. Their individual functions are becoming clear now, but an understanding of the extensive and multifaceted integration of mitochondrial and cellular activities will be a challenge for some time to come. From the perspective of a systems biologist:

A genome, organized into chromosomes that are condensed into nucleosomes, is expressed by the action of enhanceosomes, transcriptosomes, and spliceosomes as a transcriptome, and with the help of ribosomes the transcriptome is turned into a proteome. Chromosomes, consisting mostly of autosomes, but also X and Y chromosomes, are duplicated by a replisome. Members of the proteome are organized in higher-order structures such as peroxisomes, lysosomes, endosomes, and so on. Undesirable members of the proteome are attacked by proteasomes (degradomes). Syndromes arise when specific members of the proteome are absent or misbehave. A large number of metabolomes are responsible for providing the required energy and raw materials, while signalosomes or kinomes regulate the creation of order out of chaos. Under certain conditions, constituents of the signalosome activate the apoptosome to organize the return to chaos. Somewhere in all of this ***mitochondria*** *play an absolutely essential role.*

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