19.2.2 Counter-Action of Mitochondrial Dysfunction and Anti-Oxidant Actions

Numerous studies have investigated protective effects of melatonin at the level of mitochondria. Various models have followed the concept of inducing mitochondrial dysfunction by applying mitochondrial toxins, high-grade inflammation due to sepsis or endotoxemia, ischemia-reperfusion, excitotoxicity and other means of causing calcium overload, frequently with the aim of inducing apoptosis, autophagic cell death or mitophagy.[5,77] Despite the medical relevance of these approaches, they may not sufficiently cover the changes of mitochondrial function in aging, although there is certainly some overlap.

Mitochondria are protected by melatonin in multiple ways, which comprise (1) reduction of electron dissipation and, thus, free radical formation by modulating electron flux, (2) enhanced *de novo* synthesis of respirasomal proteins, (3) prevention of blockades of the electron transport chain (ETC) by reducing damage caused by oxidation, nitration and nitrosation of ETC components, (4) prevention of long-lasting opening of the mitochondrial permeability transition pore (mtPTP), (5) up-regulation of anti-oxidant enzymes, (6) improvement of the redox balance of glutathione (GSH/GSSG ratio), (7) inhibition of cardiolipin peroxidation, a crucial enzymatic step leading to dysfunction and apoptosis, (8) prevention of Ca^{2+} overload by anti-excitatory and anti-inflammatory actions, (9) increasing the number of mitochondrial DNA (mtDNA) copies, and (10) favouring the maintenance of mitochondrial mass including the inhibition of mitophagy. These numerous actions have been multiply reviewed under various aspects, including their relevance to aging.[5,11,77–85]

Instead of repeating all these details in a general context, several specific aspects of particular importance to aging shall be discussed. The free radical theory of aging assumes progressive damage to mitochondria by radicals that are largely formed in these organelles, *e.g.*, by electron dissipation from the ETC, with the consequence of increasing rates of radical production.[86–91] The primarily formed superoxide anions ($O_2\bullet^-$) can lead to free radicals of higher reactivity, either *via* H_2O_2 and the Fenton reaction or by combination with •NO to peroxynitrite ($ONOO^-$), from which hydroxyl radicals (•OH), carbonate radicals ($CO_3\bullet^-$) and $\bullet NO_2$ are formed.[11,77,79,83] The mitochondrial formation of CO_2 in the citric acid cycle may indicate an enhanced importance of $CO_3\bullet^-$ and $\bullet NO_2$ deriving from the peroxynitrite–CO_2 adduct ($ONOOCO_2^-$).[83] In comparison to •OH, the role of $CO_3\bullet^-$ may have been underrated, since it is sufficiently reactive to oxidize many biomolecules but is, by virtue of its resonance stabilization, considerably longer-lived than •OH. Another assumption of the free radical theory of aging, namely the progressive damage of mtDNA, may, however, turn out to be overrated. First, the absence of histones at mtDNA has been misinterpreted in terms of naked, unprotected DNA, although it is, in fact, densely covered by proteins such as the mitochondrial transcription factor A (mtTFA) and other integral components including anti-oxidant enzymes.[92] Second, a study on aging mtDNA mutator mice