Chapter 5
Aβ in Mitochondria—One Piece in the Alzheimer's Disease Puzzle

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Abstract Although great advances have been made in our understanding of the neurodegenerative process in Alzheimer's disease (AD), the complete picture has not emerged and there are still pieces missing. One attractive hypothesis is that mitochondrial failure is a cause of synapse loss and cognitive impairment in AD. ATP generation by mitochondria is crucial for proper synaptic function and therefore neurons are highly sensitive to mitochondrial damage potentially leading to synapse loss and cognitive dysfunction. Several evidences indicate that mitochondria are indeed damaged and dysfunctional in the AD brain; these include mitochondrial accumulation of amyloid β-peptide (Aβ), impaired brain glucose metabolism, impaired mitochondrial fusion/fission, and increased generation of reactive oxygen species (ROS). In this chapter we will focus on the role of Aβ in mitochondria and discuss mitochondrial uptake mechanisms and interactions with mitochondrial proteins. Several evidences point towards a central role of Aβ initiating mitochondrial damage and generation of ROS in turn leading to synaptic and neuronal degeneration. Therefore, it would be of high importance to develop drugs that maintain mitochondrial integrity and prevent mitochondrial failure otherwise leading neuronal dysfunction.

5.1 Introduction

Alzheimer's disease (AD) is a multifactor disorder resulting in neuronal degeneration and memory loss. The current lack of disease modifying drugs for this detrimental disorder is an increasing problem which leaves patients, relatives, caregivers, and society with an enormous burden. In order to develop such drugs it is mandatory

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D. Praticò and P. Mecocci (eds.), *Studies on Alzheimer's Disease*, Oxidative Stress in Applied Basic Research and Clinical Practice, DOI 10.1007/978-1-62703-598-9_5,

to elucidate the underlying disease mechanisms and one hypothesis that has been put forward is that mitochondrial failure is the cause of synapse loss and cognitive impairment in AD [1]. The amyloid β-peptide (Aβ) is one of the pathological hallmarks in AD and has been suggested to exert its toxicity both extra- and intracellulary [2]. Oligomeric forms of Aβ secreted from cells has for example been shown to bind to synapses and inhibit long-term potentiation [3], while intracellular Aβ accumulate in mitochondria and negatively affect mitochondrial function [4]. Aβ has been detected in mitochondria both in humans and animals [4–7]. In vitro studies show that Aβ is transported into mitochondria via the translocase of the outer membrane (TOM) machinery and localize to the mitochondrial cristae [7]. Interestingly, it has also been shown that Aβ is accumulating specifically in synaptic mitochondria in young tg AD mice [8]. In addition, a thorough study on synaptic mitochondria isolated from different brain regions from wt or AD tg mice show that hippocampal and cortical mitochondria show the highest levels of mitochondrial dysfunction (including increased ROS production and complex IV activity and decreased mitochondrial membrane potential) [9]. Together these data further support the mitochondrial hypothesis and suggest that synaptic failure detected early in the AD disease process may be caused by mitochondrial Aβ. In this chapter this hypothesis is further reviewed and mitochondrial targeting possibilities discussed. It is becoming evident that we have to treat AD early on in the disease process in order to prevent/decrease synapse loss and neurodegeneration and mitochondria emerge as one important drug target.

5.2 The γ-Secretase Complex, APP, and Aβ Are Localized to Mitochondria and Mitochondria-Associated ER Membranes

Aβ is cleaved out from the amyloid β-precursor protein (APP) by the subsequent cleavage by β- and γ-secretases. β-Secretase cleavage of APP results in the formation of a C-terminal membrane bound fragment referred to as C99. C99 is one of many substrates for the γ-secretase complex. The γ-secretase complex is membrane bound and consists of at least four different proteins, i.e., presenilin (PS1 or PS2), Nicastrin, anterior pharynx defective 1 (Aph-1), and presenilin enhancer-2 (Pen-2) [10]. APP is a type I transmembrane protein located at the plasma membrane, endosomes, Golgi network and ER with its C-terminus facing the cytosol. Depending on the exact cleavage site on the APP molecule γ-secretase complex cleavage results in different lengths of the Aβ peptide. In addition to Aβ γ-secretase cleavage of APP also generates APP intracellular domain (AICD). In the non-amyloidogenic pathway α-secretase cleaves APP within the Aβ sequence leading to generation of C83. Subsequent cleavage of C83 by γ-secretase results in production of AICD and a non-amyloidogenic p3-fragment. $A\beta_{40}$ is most abundant while the longer forms, $A\beta_{42\text{-}48}$ are more prone to aggregate and form fibrils and plaques. The longer species

are also more neurotoxic as compared to $A\beta_{40}$. Mutations linked to familiar forms of AD have been identified in APP, PS1, and PS2 and are associated with an increased $A\beta_{42}/A\beta_{40}$ ratio resulting in neurotoxicity and extensive plaque formation [11]. Aβ is generated at the plasma membrane and in the ER, Golgi, endosomal/lysosomal system following the pattern of intracellular localization of APP. Interestingly, Aβ is accumulating inside mitochondria both in human AD brain and in animal models indicating that Aβ is either produced inside mitochondria and/or taken up from the outside. In transgenic mice overexpressing mutant APP (V717/F, K670M, N671L from L Mucke) $A\beta_{40}$ and $A\beta_{42}$ start to accumulate in mitochondria from 4 months old animals before formation of plaques [5, 8]. Of particular interest is that Aβ accumulation starts in synaptic mitochondria where it causes mitochondrial dysfunction by interfering with respiratory function, mitochondrial permeability transition (mPT), and mitochondrial trafficking and transport [8]. Neurons heavily rely on oxidative phosphorylation (OXPHOS) for ATP production and a large part of this ATP is used during propagation of signals at synapses and required to drive Na^+/K^+- and Ca^{2+}-pumps. Therefore, proper mitochondrial function including ATP production is essential for synaptic function and signaling.

Whether Aβ is produced inside mitochondria or taken up from the outside is not yet fully clarified. Since both APP [12] and active γ-secretase complexes [13] have been detected in mitochondria it is theoretically possible that Aβ is produced locally in mitochondria. APP has been shown to accumulate in AD brain mitochondria via arrested import leaving a large C-terminal part outside [12, 14]. Under these circumstances APP is stuck in the mitochondrial protein import pore, consisting of the translocases of the outer (TOM) and inner membrane (TIM), causing impairment of mitochondrial function and eventually cell toxicity. The import of APP is arrested due to an acidic domain at amino acids 220–290 leaving the Aβ-region outside the import pore (Fig. 5.1). Recent data from our laboratory show that the C-terminal part of APP can be inserted into the outer mitochondrial membrane (OMM) and that the mitochondrial γ-secretase cleaves APP to generate AICD which was detected in the inter membrane space [15]. However, we detected only C83 (generated by α-secretase cleavage) and not C99 (generated by β-secretase cleavage) in the mitochondrial membrane. Subsequent γ-secretase cleavage of C83 results in formation of the p3-fragment and not Aβ formation. Thus, it is more likely that Aβ is taken up from the outside of mitochondria rather than produced inside mitochondria. Aβ coming from the outside of mitochondria could either be transported to mitochondria via vesicles [16] or produced at mitochondria-associated ER membranes (MAM) [17].

MAM is a specialized region of the endoplasmatic reticulum (ER) enriched in cholesterol and the membrane composition thus similar to lipid rafts. MAM is in contact with mitochondria and connects ER and mitochondria both physically and biochemically (Fig. 5.1). MAM has a central role in phospholipid, glucose, sphingolipid, ganglioside, cholesterol, and fatty acid metabolism and also regulates calcium homeostasis and apoptosis [18]. Interestingly, all four components in the γ-secretase as well as APP has been detected in MAM. γ-Secretase activity was

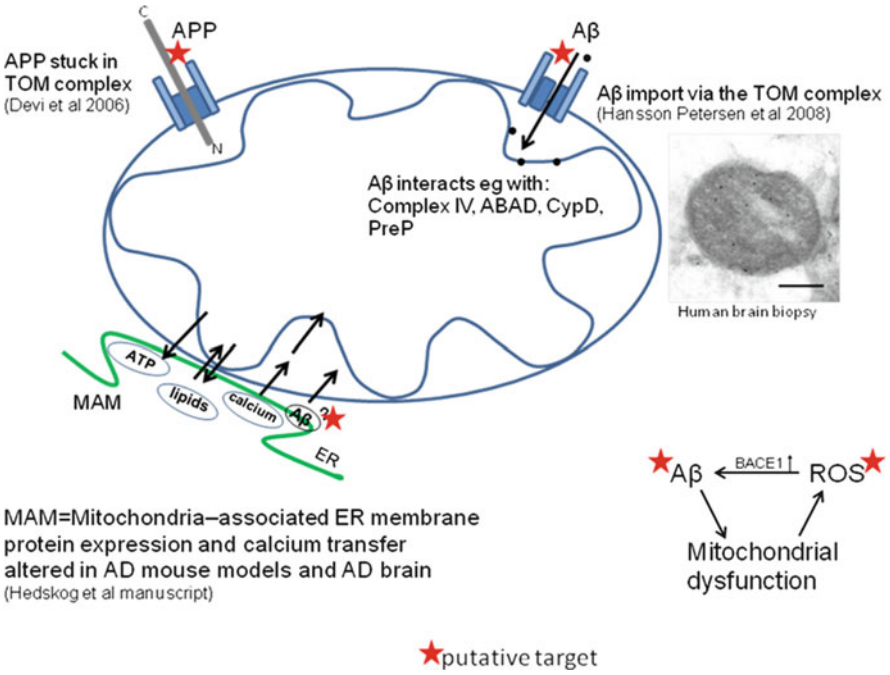

Fig. 5.1 Schematic illustration of the current knowledge of Aβ in mitochondria. APP is partly imported via the TOM complex and then stuck in the import pore causing impairment of mitochondrial function [14]. Aβ is also imported via the TOM complex and accumulate in the inner membrane where it interacts with proteins of for example the electron transport chain and mitochondrial membrane transition pore [7]. Aβ might be generated in mitochondria-associated ER membranes (MAM), where both APP and the γ-secretase complex have been detected [17], and then transported into mitochondria via TOM

detected using both a fluorescence based energy transfer-based assay and Western blotting to detect AICD [17]. The results showed that the highest γ-secretase activity were detected in MAM as compared to plasma membrane, ER and mitochondrial fractions. We have previously detected active γ-secretase complexes in mitochondria [13] and calculated that a few percent of the total γ-secretase activity in tissue is executed by the mitochondrial γ-secretase (Ankarcrona M *unpublished data*). As described above we do not have data supporting local Aβ production in mitochondria; however, we have detected AICD formation in mitochondria [15]. The function of AICD in mitochondria is presently unknown. Since rather high levels of γ-secretase activity were detected in MAM [17] it is tempting to speculate that Aβ produced at MAM can reach mitochondria via the TOM import machinery in the outer mitochondrial membrane (Fig. 5.1). This uptake mechanism was shown by in vitro import studies in isolated mitochondria performed in our laboratory [7] and further described below.

5.3 Mitochondrial Aβ Uptake Mechanisms and Submitochondrial Localization

As discussed above many studies have shown the accumulation of Aβ in mitochondria both from human AD brain and tg mutant APP mice. Several studies have also shown that Aβ cause mitochondrial toxicity and it would be presumably beneficial to block mitochondrial Aβ uptake as a treatment strategy for AD. Therefore, we undertook a study investigating the mechanisms for mitochondrial Aβ uptake (Fig. 5.1). The rationale was to use purified rat liver mitochondria treated with 0.1 μM $Aβ_{1–40}$ or $Aβ_{1–42}$ in the absence or presence of antibodies or inhibitors directed to various mitochondrial translocases, pores and channels [7]. Both $Aβ_{1–40}$ and $Aβ_{1–42}$ were taken up by mitochondria during the 30 min incubation period. The Aβ uptake was not affected by the presence of antibodies directed towards the voltage-dependent anion channel (VDAC) nor in the presence of Cyclosporine A, which is an inhibitor of the mitochondrial permeability transition pore (mPTP). In contrast, import of both $Aβ_{1–40}$ and $Aβ_{1–42}$ was prevented when import competent mitochondria were pre-incubated with antibodies directed towards proteins of the TOM complex, i.e., TOM20, TOM40, TOM70. Aβ import was not affected by the addition of valinomycin, an ionophore which cause depolarization of the mitochondrial inner membrane, indicating that the Aβ import was not dependent on the $ψ_{mit}$. After import Aβ was mostly localized to mitochondrial cristae and associated with the inner membrane fraction. It may be a hydrophobic interaction between Aβ and the TOM receptors leading to import over the outer mitochondrial membrane (OMM). Since Aβ has no classical import signaling sequence Aβ is not further imported into the matrix via the translocase of the inner membrane (TIM). It was previously reported that Aβ co-localizes with the mitochondrial matrix protein Hsp60 in mouse and human samples [5]. One explanation to the discrepancy between this and our study might be that in our in vitro assay we studied Aβ localization after 30 min of import, whereas Caspersen et al. report data from postmortem AD brains and 8-months-old transgenic APP mice. However, our data from human brain biopsies obtained from living subjects, displaying Aβ aggregates in the neuropil, show Aβ immuno-gold labeling in association with mitochondrial inner membranes [7]. Moreover, Singh et al. have in a bioinformatic study predicted that Aβ is localized to the inner membrane and rule out the presence of Aβ in the matrix [19]. Still we cannot exclude that a fraction of Aβ can be released or escapes from the membrane and into the matrix. In summary, these data show that Aβ is imported via the TOM complex where TOM 20 and TOM70 are receptors and TOM40 forms a pore in the OMM.

Recently Roses and colleagues reported that a polymorphic poly-T variant in the *TOMM40* gene (rs10524523) can be used to estimate the age of LOAD onset for *APOEε3* carriers. *APOEε3/4* carriers with very long/long poly-T repeats linked to *APOEε3* had an age of onset 7 years earlier as compared to individuals with shorter repeats [20]. *TOMM40* and *APOE* genes are separated by only ~2 kb on chromosome 19. In a novel study we investigated the effect of different poly-T lengths in

TOMM40 on the mRNA, protein and mitochondrial levels using fibroblasts from healthy *APOEε3/4* individuals carrying either short/long poly-T or very long/long poly-T (*APOEε4* always brings a long poly-T in *TOMM40*) [21]. A modified protein with potentially impaired function could for example negatively influence protein import into mitochondria which in turn would lead to mitochondrial deficiency and neuronal death explaining the earlier age of onset in *APOEε3/4* carriers with very long/long poly-T repeats. However, in our study we detected no differences in any of the parameters measured (e.g., mRNA splicing/exon skipping, TOM40 expression levels, mitochondrial membrane potential, mitochondrial area and morphology) [21]. Thus, these data, obtained from a rather limited sample set, do not support the hypothesis that the polymorphism rs10524523 directly influence the function of Tom40 and mitochondria. So far we have not investigated if the rate of mitochondrial Aβ import is affected in cells carrying this polymorphism. This may be worthwhile pursuing both for rs10524523 and other polymorphisms in *TOMM40* linked to AD.

To interfere with the TOM import machinery as a treatment strategy for AD is not trivial since this machinery cannot be blocked for import of proteins required for mitochondrial function. One possibility would be to identify the binding site for Aβ on for example TOM20 or TOM70 receptors and screen for compounds that specifically blocks this interaction without affecting import of other proteins.

5.4 Aβ Interaction with Mitochondrial Proteins

Within mitochondria Aβ has been shown to interact with several different proteins causing mitochondrial dysfunction and cell toxicity (Fig. 5.1). Here some examples of Aβ-protein interactions will be discussed (for an additional review see ref. [22]).

5.4.1 Electron Transport Chain Enzyme Complexes

Analyses of AD postmortem brain have shown decreased activity of cytochrome c oxidase (COX) also known as complex IV [23–25]. Also platelets from AD patients and AD cybrid cells have a complex IV deficiency [26, 27]. In vitro studies with mitochondria isolated from human leukocytes suggest that $A\beta_{1-42}$ inhibits complex IV activity in a copper-dependent manner [4]. The complex IV was specifically damaged in line with other studies [28–30] reporting that $A\beta_{25-35}$ selective damage complex IV and not complex I, II, or III. Crouch et al. [4] showed that low molecular weight oligomers were the toxic $A\beta_{1-42}$ species responsible for complex IV inhibition. As mentioned above complex IV activity was also shown to be decreased in mitochondria isolated from tg APP_{swe} and tg $APP_{swe}/PS1_{M146V}$ mice [9]. Further evidence for Aβ induced inhibition of complex IV activity also comes from 3× Tg-AD mice expressing mutations in APP, PS1 and Tau. In this model compromised energy production including decreased complex IV activity preceded plaque formation [31].

In another study complex IV activity was decreased in 2× APP/PS2 and 3× APP/PS2/Tau Tg AD cortices but not in mice with a Tau mutation. Instead the tau mice (pR5) had impairments in complex I activity [32]. These data confirm that it is mainly Aβ pathology that affects complex IV activity.

5.4.2 The Mitochondrial Permeability Transition Pore

Opening of the mitochondrial permeability transition pore (mPTP) results in loss of mitochondrial membrane potential, swelling of mitochondria and the release of pro-apoptotic proteins from the intermembrane space (IMS). The protein composition of mPTP has not been fully elucidated and different models have been proposed. One model is that the voltage-dependent anion channel (VDAC) in the OMM forms the pore together with adenine nucleotide translocase (ANT) and inorganic phosphate carrier (PiC) in the IMM [33]. Aβ has been shown to specifically interact with cyclophilin D (CypD), a mitochondrial matrix protein that associates with the inner membrane during opening of the mitochondrial permeability transition pore (mPTP) [34]. Translocation of matrix Cyclophilin D (CypD) to the inner membrane and CypD binding to PiC has also been proposed to trigger opening of calcium-sensitive nonspecific channels [19]. Cortical mitochondria from CypD deficient mice are resistant to Aβ- and calcium-induced mitochondrial swelling and permeability transition. Moreover, Tg mAβPP/CypD-null mice had improved learning and memory and synaptic function both in 12 and 24 months old animals [34, 35]. Aβ has also been predicted to interact with ANT in the inner membrane [19]. Simulation of protein-protein interactions suggested that the ANT-Aβ interaction is stronger that the CypD-Aβ interaction. At present it is not known what function the ANT-Aβ interaction has; however, it may affect the normal physiological function of ANT which is transport of ATP and ADP.

5.4.3 Mitochondrial Aβ-Binding Alcohol Dehydrogenase and Presequence Protease

Two different Aβ-binding proteins have been identified in the mitochondrial matrix, i.e., mitochondrial Aβ-binding alcohol dehydrogenase (ABAD) and Presequence Protease (PreP). Our data as described above show that Aβ is located to the inner membrane after import via the TOM40 pore. To what extent this Aβ fraction is available for ABAD and Prep P interactions in the matrix is not clear at present. ABAD has been found to be up-regulated in neurons from AD patients [36] and Aβ has been shown to interact with ABAD resulting in free radical production and neuronal apoptosis. ABAD was identified as an Aβ-binding protein in a yeast two-hybrid screen [36]. ABAD is localized to the mitochondrial matrix and has an essential physiological role in mitochondria. ABAD-Aβ complexes were detected in AD brain and in Tg mutant AβPP/ABAD (Tg mAβPP/ABAD) mice. Cortical

neurons cultured from Tg mAβPP/ABAD mice show increased production of ROS and decreased mitochondrial membrane potential, ATP levels, and activity of respiratory chain complex IV. Consistently, these neurons displayed DNA-fragmentation and caspase-3 activity resulting in cell death by day 5–6 in culture [37]. ABAD uses NAD^+ and/or NADH as its cofactor and catalyzes the reversible oxidation and/or reduction of alcohol group in its substrates [38]. The crystal structure of ABAD-Aβ complexes has been determined showing that the NAD^+ binding pocket is distorted, hindering NAD^+ from binding to ABAD in the presence of Aβ [36, 38]. Thus, Aβ blocks ABAD activity causing mitochondrial dysfunction and ultimately cell death. Two stretches of ABAD residues in the L_D loop region (amino acids 95–113) have been shown to be important for Aβ binding. Cell permeable peptides ABAD-DP (ABAD-decoy peptide fused to the Tat protein and a mitochondrial targeting signal) administrated to transgenic APP mice blocked formation of ABAD-Aβ complexes in mitochondria, attenuated oxidative stress, increased mitochondrial respiration, and also importantly improved spatial memory [39]. Thus, the use of inhibitors of ABAD-Aβ interaction emerges as a novel therapeutic strategy for AD.

PreP is also localized to the mitochondrial matrix and putatively responsible for the degradation of the accumulated Aβ in mitochondria [40]. PreP was originally found and characterized in *Arabidopsis thaliana* [41] as a protease degrading targeting peptides that are cleaved off in mitochondria after completed protein import as well as other unstructured peptides up to 65 amino acid residues in length, but not small proteins [42, 43]. Recombinant hPreP completely degrades both Aβ40 and Aβ42 as well as Aβ Arctic protein (42, E22G) at unique cleavage sites including several sites in a very hydrophobic C-terminal Aβ (29–42) segment that is prone to aggregation. Interestingly, PreP is an organellar functional analogue of the human Insulin Degrading Enzyme (IDE), implicated in AD as it cleaves Aβ before insoluble amyloid fibers are formed [44–46]. A recent study using human and transgenic mouse brain show that PreP activity is reduced in human postmortem AD brain (temporal lobe) and in mice overexpressing mutant AβPP (m AβPP, J-20 line) or mutant AβPP together with ABAD (AβPP/ABAD) [47] Enhanced production of ROS may cause the decreased PreP proteolytic activity resulting in mitochondrial Aβ accumulation and in turn leading to toxicity and neuronal degeneration. Interestingly, it has also been shown that the Aβ degrading enzymes neprilysin and IDE are subject to oxidative inactivation [48]. Decreased degradation of Aβ in combination with ROS induced BACE1 activity [49], as discussed below, would result in increased Aβ generation and accumulation.

5.5 The Vicious Cycle of Aβ and ROS Generation: A Putative Target?

So far we have discussed data showing that Aβ can enter mitochondria, bind to various proteins and thus induce for example ROS production or disruption of mitochondrial integrity. However, in a recent publication Leuner et al. [49] show that cells treated with toxins (i.e., rotenone and antimycin) inhibiting respiratory

complex I and III respectively and subsequently inducing ROS production trigger upregulation of BACE1 activity and increased secretion of Aβ. Also animals with deficiency in complex I (Ndufs3 KO mice) show high production of ROS and increased levels of secreted Aβ. These data suggest that dysfunction in the respiratory chain trigger an increased generation of Aβ. The secreted Aβ may then be toxic by binding to synapses or internalized and for example transported into mitochondria where it further impairs respiratory function initiating a vicious cycle of ROS and Aβ generation (Fig. 5.1). Mitochondria are the main source of ROS in the cell and these free radicals can affect targets (proteins, lipids, RNA, DNA) both inside and outside mitochondria. The study by Leuner et al. [49] reinforces the importance of controlling ROS production in cells as one possible treatment for AD. Indeed it was recently published that MitoQ a mitochondria targeted antioxidant had positive effects on cognition in mice after 4–5 months treatment [50]. MitoQ is accumulating 500–1,000× inside mitochondria and efficiently scavenging ROS at the spot of its production. MitoQ has been through two phase II trials for Parkinson's disease [51]. For an extended review of other antioxidants tested as AD treatment [52]. No matter what is "the hen or the egg" selective modulation of BACE1 and/or γ-secretase activity and antioxidants targeted to mitochondria are two treatment strategies worth pursuing in order to maintain proper mitochondrial function and synapse activity.

At present Alzheimer's disease researchers are questioning what went wrong when the clinical trials of different compounds and antibodies designed to interfere with Aβ production/aggregation/clearance failed. The common sense is that (a) inhibitors of γ-secretase are not useful; instead we need molecules that modulates γ-secretase activity specifically towards APP (b) BACE1 is a difficult target (c) we need to enrol patients who are in early stages of the disease. The last point put high demands on reliable diagnosis with a combination of validated biochemical biomarkers, brain imaging and neuropsychological testing. Most clinical trials conducted so far have enrolled patients with mild to moderate or even severe AD and no consistent improvement of cognition has been reported. Probably these patient's neurons have already started to degenerate in high degree and even though data show a decreased plaque burden the neurons cannot be rescued at this late stage. Our own experience from the Latrepidine (Dimebon) study points in the same direction: to be efficient treatment has to be given before mitochondria and other cell functions are damaged.

Dimebon was originally approved in the former Soviet Union as a nonselective antihistamine for skin allergy and allergic rhinitis [53], but was withdrawn from the market with the advent of more selective treatments. Dimebon attracted renewed interest due to findings suggesting a neuroprotective effect [54–56]. In a Phase II AD trial, dimebon treatment was associated with benefits on cognition, global function, activities of daily living, and behavior [57]. Several Phase III clinical trials were then performed for both AD and Huntington's disease [58]. However, all clinical trials with dimebon have now been terminated since no positive effects of the drug treatment were obtained.

Dimebon exhibits a rich pharmacological profile and binds to histamine-, adrenergic-, dopamine-, and serotonin-receptors [56, 59]. It is known to be a weak inhibitor of: acetylcholinesterase ($IC_{50} = 8–42$ μM) [55], N-methyl-D-aspartate (NMDA) receptors ($IC_{50} = 10$ μM) [56, 60], and voltage-gated calcium channels

(IC_{50} = 50 μM) [56, 61]. In addition, μM concentrations of dimebon have previously been shown to protect against neuronal cell death induced by $A\beta_{25–35}$ [55] and to modulate the mitochondrial permeability transition pore (10–200 μM) [62]. In a study from our laboratory [63] we show that nM concentrations of dimebon (1–5 days incubation) results in an increase of mitochondrial membrane potential (hyperpolarization) and cellular ATP levels both in mouse cortical neurons and human neuroblastoma cells. Moreover, dimebon pretreatment made cells more resistant to depolarization of mitochondrial membrane potential induced by high intracellular calcium concentrations. Cells were also protected from undergoing cell death induced either by calcium stress or withdrawal of growth factors. Our study suggests that dimebon directly or indirectly affects mitochondria making cells more resistant to cell death stimuli. Based on our in vitro data it is still possible that dimebon might work more efficiently if given to patients early in the disease process.

5.6 Conclusions

Accumulating evidence both from human brain as well as AD animal and cell models show that Aβ is imported into mitochondria where it accumulates in the inner membrane and bind to various proteins causing mitochondrial failure and cell toxicity. The consistent inhibition of complex IV (COX) in the respiratory chain by Aβ has been shown by several laboratories. Interestingly, in a recent study [49] it was shown that complex I inhibition or deficiency resulting in increased generation of ROS leads to activation of BACEI and increased secretion of $A\beta_{40}$. Together the data suggests a vicious cycle of ROS and Aβ generation, mitochondrial failure and neuronal degeneration. Importantly, accumulation of Aβ appears to be an early event during the disease process, as Aβ for example has been shown to first accumulate in synaptic mitochondria in young AD tg mice [8]. It is therefore possible that mitochondrial Aβ accumulation is one cause of synaptic failure correlating with cognitive impairment in AD. Future treatment strategies should take this into account and drugs targeting mitochondria developed.

Note added in proof For a recent publication about the role of ER-mitochondria interplay in AD see: Modulation of the endoplasmic reticulum-mitochondria interface in Alzheimer's disease and related models. Hedskog L, Pinho CM, Filadi R, Rönnbäck A, Hertwig L, Wiehager B, Larssen P, Gellhaar S, Sandebring A, Westerlund M, Graff C, Winblad B, Galter D, Behbahani H, Pizzo P, Glaser E and Ankarcrona M. Proc Natl Acad Sci USA. 2013;110:7916–21.

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