

# Molecular & Cellular Therapies



***Molecular and Cellular Therapies*** (2018) 6:1

The role of mitochondria-targeted antioxidant MitoQ  
in neurodegenerative disease

Linlin Zhang, Aurelio Reyes, Xiangdong Wang

## Review

# The role of mitochondria-targeted antioxidant MitoQ in neurodegenerative disease

Linlin Zhang<sup>1</sup>, Aurelio Reyes<sup>2</sup>, Xiangdong Wang<sup>3</sup>

<sup>1</sup>The First Affiliated Hospital of Wenzhou Medical University, Wenzhou, China;

<sup>2</sup>MRC Mitochondrial Biology Unit, University of Cambridge, Cambridge CB2 0XY, United Kingdom;

<sup>3</sup>Zhongshan Hospital Institute of Clinical Science, Fudan University Medical School, Shanghai, China

Correspondence to: Xiangdong Wang, MD, PhD, Prof, E-mail: Xiangdong.wang@clintransmed.org

### Abstract

The discovery of charged molecules being able to cross the mitochondrial membrane has prompted many scholars to exploit this idea to find a way of preventing or slowing down aging. In this paper, we will focus on mitochondria-targeted antioxidants, which are cationic derivatives of plastoquinone, and in particular on the mitochondria-targeted antioxidant therapy of neurodegenerative diseases. It is well known that the accumulation of amyloid- $\beta$  peptide (A $\beta$ ) in mitochondria and its related mitochondrial dysfunction are critical signatures of Alzheimer's disease (AD). In another neurodegenerative disease, Parkinson's disease (PD), the loss of dopaminergic neurons in the substantia nigra and the production of Lewy bodies are among their pathological features. Pathogenesis of Parkinson's disease and Alzheimer's disease has been frequently linked to mitochondrial dysfunction and oxidative stress. Recent studies show that MitoQ, a mitochondria-targeted antioxidant, may possess therapeutic potential for A $\beta$ -related and oxidative stress-associated neurodegenerative diseases, especially AD. Although MitoQ has been developed to the stage of clinical trials in PD, its true clinical effect still need further verification. This review aims to discuss the role of mitochondrial pathology in neurodegenerative diseases, as well as the recent development of mitochondria targeted antioxidants as a potential treatment for these diseases by removing excess oxygen free radicals and inhibiting lipid peroxidation in order to improve mitochondrial function.

### Keywords

Alzheimer's disease, mitochondria-targeted antioxidant, MitoQ, oxidative stress, Parkinson's

disease

### Introduction

Currently there are three pharmacological therapy approaches for mitochondrial disorders [1]: the first approach is to engineer compounds which selectively target to mitochondria; the second one is to design compounds that are not targeted to mitochondria but act on them based on their binding to specific targets; and the third one is to use compounds located outside the mitochondria but that ultimately affect the function of mitochondria [2]. Mitochondria-targeted antioxidants, 10-(6'-plastoquinonyl) decyl-triphenylphosphonium (SkQ1) and its analog plastoquinonyl decylrhodamine 19 (SkQR1), have been proved to slow down or even reverse the effects of aging [3, 4, 5]. Another important antioxidant is MitoQ, a synthetic analog of coenzyme Q10. As a mitochondria-targeted antioxidant, MitoQ can selectively accumulate in mitochondrial and remove excess oxygen free radicals, prevent lipid peroxidation and protect mitochondria and cell membranes against oxidative stress. An ever-increasing number of studies suggest that MitoQ possess some positive effect in treating neurological diseases with mitochondrial dysfunction and oxidative stress as main pathological features. In particular, the pathogenesis of neurodegenerative diseases, Alzheimer's and Parkinson's disease, is very complex because of the involvement a vast number of factors. Since mitochondrial dysfunction and oxidative stress are among the most relevant factors [6] and they happen in the early onset of neurodegenerative diseases, they can be considered as essential and potential therapeutic targets for the early treatment of the disease.

### Neurodegenerative diseases

Alzheimer's disease is a worldwide neurodegenerative disease typically correlated with aging. About 36.5 million people around the world are affected and this

number will rapidly increase in the future, especially in China, India, and Latin America [7]. In addition, young-onset cases have more frequently been reported and to date there is no effective treatment that can be used as a cure. This disease is characterized by wide spread synaptic damage and neuronal death with accumulation of intracellular neurofibrillary tangles and extracellular amyloid plaques of mainly A $\beta$  [8]. In the early stages of AD, mitochondrial dysfunction induced oxidative stress can result in macromolecule damage, mainly lead by hydroxyl radicals [9]. The different aspects of the disease have been revealed through the studies on Alzheimer cells and transgenic mouse models which allow the direct observation of the pathogenic mechanisms of AD, especially the interaction between A $\beta$  and mitochondria. Mitochondria are not only the target for amyloid precursor protein (A $\beta$ PP) that releases and accumulates A $\beta$  in the mitochondrial import channels, but also for A $\beta$  which interacts with different proteins in the mitochondria, affecting normal mitochondrial function [10]. In humans, A $\beta$ PP is present in many cellular structures like plasma membrane and many organelles such as endoplasmic reticulum, Golgi apparatus, as well as mitochondria. A $\beta$ PP produces A $\beta$ , a peptide of 39-43 amino acids in length, via the action of a  $\beta$ -secretase and then a  $\gamma$ -secretase enzyme [11]. Mitochondria, endosomes, Golgi and endoplasmic reticulum have been described as the major targets of A $\beta$ . In mitochondria, A $\beta$  can induce dysfunction of mitochondrial Ca<sup>2+</sup> channels [12] and affect the activity of mitochondrial. Indeed, pyruvate dehydrogenase and  $\alpha$ -ketoglutarate dehydrogenase can be altered decreasing NADH reduction, as well as the electron transport chain (ETC) enzyme complex IV, causing a reduction in the proton pumping across the inner mitochondrial membrane and hence altering the mitochondrial membrane potential [13]. The effect on complex V activity results in a decrease in ATP production along with an increasing in reactive oxygen species (ROS) production (Figure 1). Normally, excess ROS is removed by the antioxidant defense system, but oxidative stress may lead to a series of injuries due to an imbalance of antioxidant and prooxidant homeostasis, such as the damage of nucleic acids, proteins, and lipids [14]. On the other hand, excessive ROS negatively regulates presequence P (PreP) activity, hindering the normal degradation of A $\beta$  and leading to an increase in A $\beta$  content in mitochondria. Additionally, ROS induces mitochondrial peroxidation of macromolecules such as mitochondrial DNA and lipids, and it also damages the mitochondrial function by interfering with the fusion and fission processes of mitochondria, leading to mitochondrial fragmentation [15] and increased mitochondrial DNA mutations [10]. Besides, excess production of ROS also induces synaptic dysfunction by increasing tau hyperphosphorylation and neurofibrillary tangle formation, eventually leading to synaptic failure. There is wide evidence that oxidative stress plays an essential role in the pathological process of AD induced by A $\beta$ . Therefore, it sounds to work on the elimination of free radicals and prevent them from being produced as a therapy for AD. In

fact, many studies have been carried out with this scope. In one study, a mitochondria-targeted antioxidant, MitoQ, has been used to reverse the cognitive decline and to prevent early neuropathology in a transgenic mouse model of AD by acting against the attack of A $\beta$ -induced toxicity and oxidative stress [16].

Parkinson's disease is the second prevalent progressive neurodegenerative disease that affects about 2% of people over the age of 60 [17]. One of the pathological characteristics of PD is the loss of dopaminergic neurons in the substantia nigra pars compacta that causes motor symptoms. The other feature is the dopamine depletion in the striatum. PD is also associated with the presence of Lewy bodies in surviving dopaminergic neurons [18]. To date, the treatment of PD remains only on the surface, and current therapies cannot effectively slow or prevent the pathologic progression of the disease [19]. Although extensive studies have been carried out on PD, its precise etiology remains largely unknown despite some pathological mechanisms, such as mitochondrial dysfunction, oxidative stress and the ubiquitin-proteasome system disruption, as well as neuroinflammation have been brought forward. Under these assumptions, oxidative stress and mitochondrial dysfunction could be considered as reasonable candidates. In fact, mitochondrial DNA mutations have been shown to play a significant role in the death of dopaminergic neurons [20] and a great wealth of evidence suggests that mitochondrial dysfunction plays a vital role in the course of PD pathogenesis. One of the most significant research lines is the study of MPTP (1-methyl-4-phenyl-1, 2, 3, 6-tetrahydropyridine), a Parkinsonian toxin which selectively depressed mitochondrial complex I inducing Parkinsonian syndromes [21]. Another toxic metabolite, originated by the action of monoamine oxidase on MPTP is 1-methyl-4-phenylpyridinium (MPP<sup>+</sup>) [22]. It is transported into the dopaminergic neurons by the dopamine transporter and accumulates in the mitochondria, causing adenosine triphosphate depletion through inhibition of complex I activity, changes in mitochondrial membrane potential, increases in the production of ROS and finally leads to apoptotic cell death [23]. Similarly, other complex I inhibitors for like rotenone, fenpyroximate and trichloroethylene also lead to dopaminergic neurodegeneration in many research models include human, all indicating mitochondrial dysfunction plays an important role in PD pathomechanism [24]. These toxic substances affect the normal functioning of mitochondria, result in a decreasing of function of electron transport chain [25] and mitochondrial mobility [26]. Furthermore, they can also induce an increase of the mitochondrial permeability transition and the production of ROS, as well as an increase in the activity of nitric oxide synthase. In PD patients, the function of complex I is impaired in the substantia nigra. The change in complex I structure is due to the absence of apoptosis inducing factors which does not cause neurodegeneration due to the loss of dopaminergic neurons, but makes the dopaminergic neurons more susceptible to neurotoxins

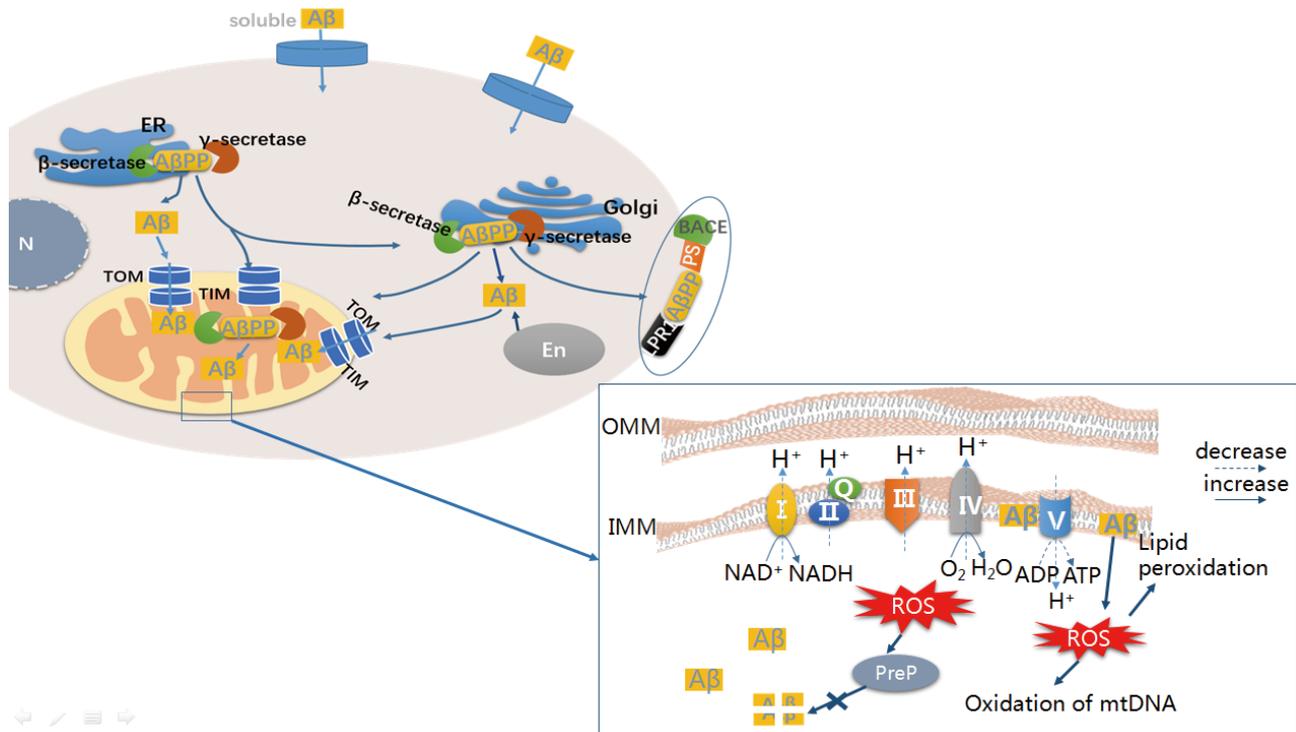


Figure 1. The generation and influence of Aβ. AβPP is synthesized in the endoplasmic reticulum (ER) and then directed to Golgi network, the cell surface or to mitochondria. In the plasma membrane, the apolipoprotein receptor LRP1 forms a complex with AβPP, together with other plasma membrane enzymes, such as the β-secretase BACE and the γ-secretase presenilin (PS), inducing the internalization of the amyloid precursor protein. AβPP produces Aβ, a 39-43 amino acid polypeptide via the activity of β-secretase and then γ-secretase enzymes [11]. Aβ originates in the endosome (En), Golgi and ER, and it is especially present in Golgi or in late endosomes following the reuptake from the cell surface. Aβ enters into the mitochondrial matrix through TOM and TIM or is derived from mitochondria-associated AβPP metabolism. In mitochondria, Aβ can affect several enzymes inducing NADH reduction and the ETC enzyme complex IV, causing a reduction in the amount of protons and resulting in a decrease in ATP, which increases ROS production. Excessive amounts of ROS leads to lipid peroxidation and negatively regulates presequence P (PITM1/PreP) activity, hindering the normal degradation of Aβ and leading to an increase in Aβ content in mitochondria.

[27].

Pathological mutations in many genes such as Parkin, α-synuclein, LRRK2 and DJ-1, PINK1, directly or indirectly demonstrate that mitochondrial dysfunction plays a role in PD patients [17]. Parkin, the E3 ubiquitin ligase, has been associated with the function of the respiratory chain. A lack of Parkin in mice and flies, for instance, would show a reduction in the level of proteins involved in mitochondrial function, reduced activity of complex I and complex IV of the electron transport chain and disrupted mitochondrial integrity [28]. Mutations in Parkin cause autosomal recessive juvenile PD. Similarly, mutations in PINK1 (PTEN-induced putative kinase 1) can also lead to an autosomal recessive form of PD, and such form is familial and early-onset. In cell culture models of Parkinsonism, mutations in PINK1 result in decreasing mitochondrial respiratory chain function, leading to reduced ATP synthesis and the increase of α-synuclein aggregation [29]. PINK1 localizes to mitochondria via a multisubunit complex that includes the outer membrane protein TOM40 and

the inner membrane protein TIM23. Upon translocation, it is cleaved by MPP and PARL proteases leading to its degradation. However, under stress conditions or in damaged mitochondria, it cannot be cleaved and stabilizes in the outer membrane leading to autophosphorylation and serine phosphorylation of ubiquitin and Parkin, promoting ubiquitination of the mitochondrial outer membrane protein to trigger selective mitochondrial autophagy (Figure 2) [30-32]. Parkin and PINK1 knockouts in *Drosophila*, has also shown that they are on same functional pathway, and that PINK1 has the ability to upregulate Parkin [30]. Thus, pathological changes in Parkin and PINK-1 disrupt normal mitochondrial function and well-balanced mitophagy. The mutation of DJ-1 lead to a rare and early-onset form of Parkinsonism, such form of PD is also autosomal recessive. Human who carry this gene mutation exhibit mitochondrial damage to the oxidative respiratory chain in mitochondria, decrease membrane potential, and increased levels of ROS in mitochondria, as well as altered regular mitophagy [33].

To date, no specific drug has been developed to slow down the progression of PD. Consequently, new therapies are urgently needed. Coenzyme Q, a vital antioxidant in the respiratory chain, is located in the lipid layer of mitochondria. Coenzyme Q10 possess neuroprotective effects that could protect against mitochondrial toxins, for example, MPTP. Although this antioxidant has been shown to be safe and patient tolerant, it has not shown any clinical value in the phase III of clinical trials that have already been carried out [34]. MitoQ is another powerful mitochondria-targeted antioxidant, which accumulates in the mitochondria and resists the damage induced by oxidative stress. However, a study using MitoQ did not revealed a slowdown in the progression of PD [35].

### Mitochondria-targeted antioxidant therapy

The two most common approaches for the use of small molecules *in vivo* as mitochondrial targeted antioxidants are: conjugation to a lipophilic cation or absorption into mitochondria-targeted peptides.

Lipophilic cations can readily pass through the phospholipid bilayer, such as plasma membrane or the mitochondrial membrane, due to the effective distribution of charge cations in a large and hydrophobic surface that reduces their activation energy and enables their passage across the membrane [36]. These lipophilic cations can cross lipid bilayers without the need of ionophores or carrier proteins and accumulate in the mitochondrial matrix thanks to the negative-inside mitochondrial membrane potential. Triphenylphosphonium (TPP) cation is the most widely used lipophilic cation to transfer antioxidants into mitochondria. It consists of an intermediate positive charge of phosphorus and a surrounding hydrophobic surface. Therefore, TPP can quickly cross the phospholipid bilayer and retain the positive charge. Such positive charge is used to drive TPP cation first into the cell and subsequently into the mitochondrial matrix. The plasma membrane potential is 30-60 mV (negative inside), which favors a 10-fold higher concentration of TPP in the cytoplasm than outside the cell. Similarly, the much greater mitochondrial membrane potential, ~140-160mV, strengthens the reallocation of lipophilic cations from

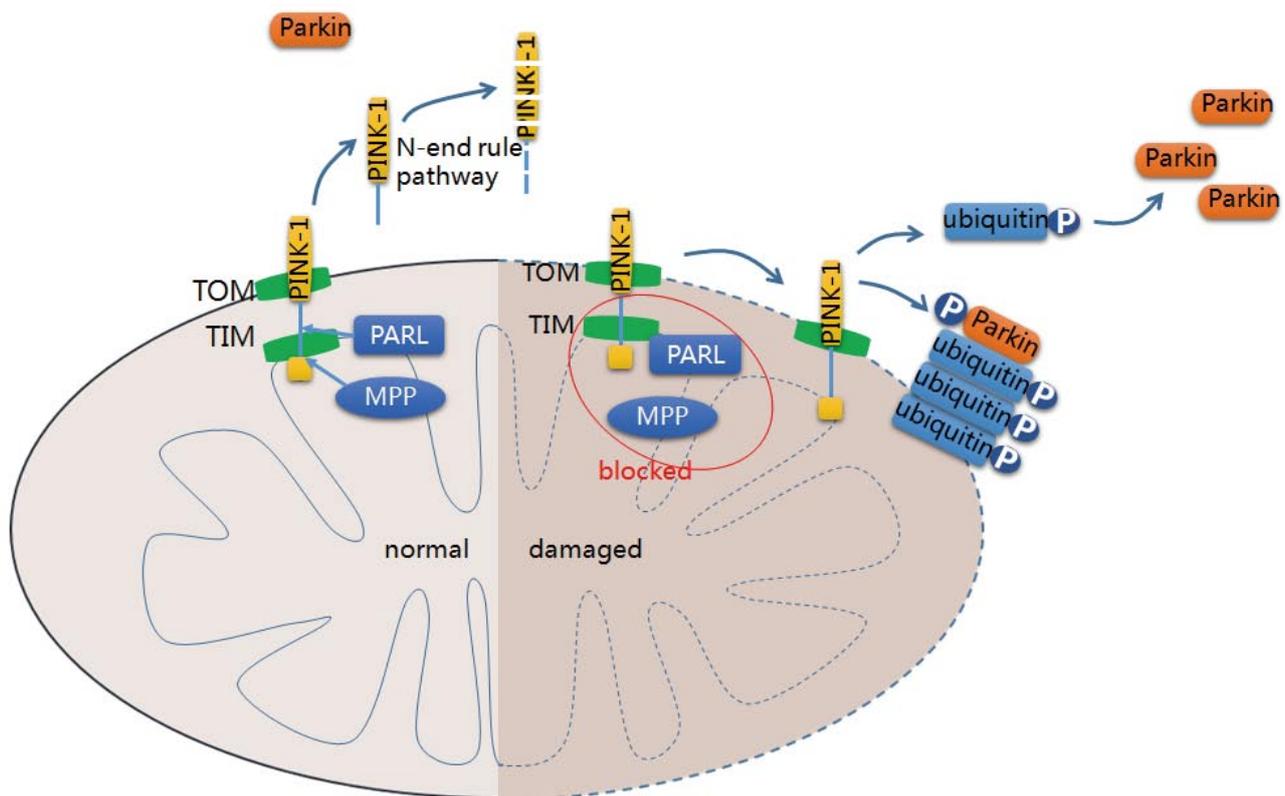


Figure 2. The PINK1–Parkin pathway. In the normal conditions, PINK1 is imported to the mitochondria through TOM and TIM complex, on the outer mitochondrial membrane (OMM) and the inner mitochondrial membrane (IMM), respectively. After entry, PINK1 is sequentially cleaved by inner mitochondrial membrane proteases MPP and PARL, between amino acids 103 and 104. The remaining N-terminal fragment is then degraded via the N-end rule pathway in the cytosol. However, when mitochondria are damaged or the TIM complex is non-functional due to mitochondrial depolarization, PINK1 processing by MPP and PARL is restrained because of lack of import into the inner membrane. Instead, PINK1 accumulates on the OMM bound to the TOM complex, where it phosphorylates ubiquitin at Ser65. This stimulates the recruitment of cytosolic Parkin to the mitochondrial surface via the direct binding to phospho-ubiquitin chains. Parkin is then also phosphorylated by PINK1 and activates it.

the intracellular space into the mitochondria, leading to ~200-400 fold accumulation of TPP in the mitochondrial matrix [37]. Based on this principle, researchers targeted many antioxidants to mitochondria by conjugation to the TPP cation, including MitoQ.

MitoQ is a mitochondria-targeted ubiquinol, it consists of an ubiquinone moiety covalently attached to a TPP moiety by a ten-carbon aliphatic carbon chain [38]. The TPP moiety on MitoQ can cross the plasma membrane due to the plasma membrane potential and then accumulate hundreds-fold in the mitochondrial matrix compared to cytosol due to its greater membrane potential [16]. Once MitoQ accumulates in the matrix side of the inner membrane, it persistently removes peroxy radicals, peroxynitrite, superoxide, prevents lipid peroxidation as well as mitochondrial injury [39, 40]. After detoxifying oxidants, the ubiquinol is oxidized to ubiquinone and recycled by the electron transport chain complex II [41, 42]. Consequently, this directionally accumulates MitoQ in the mitochondrial matrix where it is continually recycled, making this targeted-antioxidant hundreds-fold more powerful than other antioxidants [38]. On the other hand, the ubiquinol moiety of MitoQ can also react directly with superoxide and protect against peroxynitrite [43]. However, in some cases, due to the redox cycle of quinone, it may be pro-oxidant and pro-apoptotic and generate superoxide [44]. MitoQ has been shown to possess a protective effect in a large number of animal models of diseases related to oxidative stress such as neurodegenerative disease (including Alzheimer's disease, Parkinson's disease etc.) [16, 20], ischemia-reperfusion [45], type 2 diabetic [46] and hypertension [47].

To evaluate the potential effect of MitoQ on Alzheimer's disease, different mouse models were analyzed and it has been reported that MitoQ was able to prevent the accumulation of A $\beta$ , inhibit A $\beta$  induced oxidative stress and reverse the reduction of synapses and early cognitive decline. In particular, MitoQ protected neurons against A $\beta$  in a transgenic mouse model of AD and diminished the neurological deficits in the mice [16]. In cultured mouse sympathetic neurons, deprivation of nerve growth factor induced cell death but MitoQ treatment blocked apoptosis by preventing increased mitochondria-derived ROS and subsequent cytochrome c release, caspase activation, and mitochondrial damage by scavenging mitochondrial superoxide [48]. Several studies have shown that MitoQ can not only increase life span, postpone A $\beta$  induced paralysis, but also improve mitochondrial lipid cardiolipin depletion, as well as protect the ETC enzymes complex IV and complex I [49]. In SH-SY5Y cells, it could effectively inhibit mitochondrial fragmentation caused by 6-hydroxydopamine [50]. In another study, MitoQ effectively inhibited neurotoxicity induced by both MPP<sup>+</sup> and MPTP in cell cultures and in a preclinical PD mouse model [51]. MitoQ inhibited the decrease in dopamine and tyrosine hydroxylase levels, as well as the loss of mitochondrial membrane potential caused by MPP<sup>+</sup>.

It could also alleviate the depletion of dopamine and its metabolites caused by MPTP toxicity. In PD animal models, MitoQ decreased caspase-3 activation which was induced by MPP<sup>+</sup> and alleviated mitochondrial aconitase inactivation which was mediated by MPTP, maintaining normal operational tricarboxylic acid cycle, and decreasing oxidative damage [51]. In one study, a PD mouse model was treated with high levels of MitoQ for up to 28 weeks and showed no evidence of toxicity on whole-body physiology. This result indicated that mitochondria-targeted antioxidant, MitoQ, can be safely administered long-term in mice [52]. MitoQ has undergone phase I and II clinical trials in Hepatitis C virus patients and was proved to be an effective strategy, demonstrating the potential therapeutic efficacy of MitoQ in chronic liver diseases [53]. However, in another double-blind clinical trial of PD patients, it failed to demonstrate any benefit in slowing down the pathologic process of PD during 12 months [35]. A potential explanation for these conflicting results is that the extent of dopamine neurons damage has exceeded the neuroprotective effect of MitoQ. In general, approximately fifty percent of dopamine neurons have been lost by the time there is a clinical diagnosis of PD. Thus, it is possible that the treatment on the remaining neurons is no longer able to prevent the progression of the disease. Although the beneficial effects of MitoQ in PD have been proved both *in vitro* and *in vivo*, further detailed studies are needed to test and verify its potential for treatment. However, it is not only neurodegenerative diseases the targets of MitoQ therapies. In 2015, a new clinical trial aimed to study the effects of MitoQ on improving physiological functions including vascular, motor, and cognitive in middle aged and elderly people was registered on clinicaltrials.gov [54]. In a recent study on human skeletal muscle feed arteries from 44 subjects, it has been shown that MitoQ could reverse age-related endothelial vascular dysfunction [55]. Furthermore, MitoQ could reduce the severity of renal damage in renal ischemia-reperfusion injury in rat models [45]. All this suggest that mitochondrial targeted antioxidants, such as MitoQ can be applied as treatment to a wide variety of diseases.

## Conclusion

In neurodegenerative diseases, the two main pathological markers are mitochondrial dysfunction and oxidative stress. In addition to these two hallmarks, there are other pathological features, such as excessive production of ROS, mitochondrial DNA mutations or impaired ATP synthesis. Mitochondrial dysfunction plays a central role in AD and PD, and can be an important target for treating these diseases. As a new therapeutic method, mitochondria-targeted antioxidants have good pharmacological effects in various animal and cell culture models. Among them, MitoQ has been the most extensively studied. In the phase II clinical trial on PD patients, its beneficial effect has not been proven though. This may be due to the fact that mitochondrial dysfunction may represent early stages of the pathological process

of the disease, while pharmacological effects are usually pursued at later stages of the disease after the clinical diagnosis. Mitochondrial dysfunction and oxidative stress can not only define these neurodegenerative diseases, but may also be a new therapeutic strategy and therefore a bigger effort should be made to push research forward before these diseases encroaching on more people.

### Future outlook

So far, MitoQ, is the most studied mitochondrial-targeted antioxidant, which has already been used in human clinical trials. In human phase I clinical trials, MitoQ performed good pharmacokinetic characteristics when first administered to Parkinson patients to see if it could slow the progression of the disease [35]. Although mitochondrial targeted therapies inaugurated a new era in mitochondrial dysfunction, such procedure still has some unresolved limitations. First, these specific compounds are often required to be located in the mitochondrial matrix or the matrix-facing surface of the inner membrane. Second, these compounds are not yet targeted to specific organs and thus, they are commonly concentrated in organs with higher mitochondrial content. Moreover, mitochondria-targeted antioxidant MitoQ, has been shown to be safe for humans but still requires further clinical trials. The field of therapeutic mitochondrial compounds has only just begun, and there are many diseases caused by mitochondrial damage such as ischemia/reperfusion injury, diabetes and metabolic syndrome already waiting for this technology to mature. However, different diseases have diverse pathological features, which in turn, means further and specific testing.

### Abbreviations

A $\beta$ : amyloid beta

AD: Alzheimer's disease

PD: Parkinson's disease

SkQ1: 10-(6'-plastoquinonyl) decyltri-phenylphosphonium

SkQR1: plastoquinonyldecylrhodamine 19

A $\beta$ PP: amyloid precursor protein

ROS: reactive oxygen species

NADH: nicotinamide adenine dinucleotide

ETC: electron transport chain

MPTP: 1-methyl-4-phenyl-1, 2, 3, 6-tetrahydrodipyridine

MPP+: 1- methyl-4-phenylpyridinium

LRRK2: leucine-rich repeat kinase 2

DJ-1: Parkinson disease protein7

PINK-1: (PTEN)-induced putative kinase 1

TOM: translocase of outer membrane

TIM: translocase of inner membrane

TPP: triphenylphosphonium

ER: endoplasmic reticulum

PS: presenilin

En: endosome

PreP: presequence P

OMM: outer mitochondrial membrane

IMM: inner mitochondrial membrane

### References

- Smith RA, Hartley RC, Murphy MP. Mitochondria-targeted small molecule therapeutics and probes. *Antioxid Redox Signal*. 2011 Dec 15;15(12):3021-38.
- Murphy MP, Smith RA. Targeting antioxidants to mitochondria by conjugation to lipophilic cations. *Annu Rev Pharmacol Toxicol*. 2007 47:629-56.
- Shabalina IG, Vysokikh MY, Gibanova N, Csikasz RI, Edgar D, Hallden-Waldemarson A, Rozhdestvenskaya Z, Bakeeva LE, Vays VB, Pustovidko AV, et al. Improved health-span and lifespan in mtDNA mutator mice treated with the mitochondrially targeted antioxidant SkQ1. *Aging (Albany NY)*. 2017 Feb 15;9(2):315-339.
- Skulachev VP, Anisimov VN, Antonenko YN, Bakeeva LE, Chernyak BV, Elichev VP, Filenko OF, Kalinina NI, Kapelko VI, Kolosova NG, et al. An attempt to prevent senescence: a mitochondrial approach. *Biochim Biophys Acta*. 2009 May;1787(5):437-61.
- Skulachev MV, Antonenko YN, Anisimov VN, Chernyak BV, Cherepanov DA, Chistyakov VA, Egorov MV, Kolosova NG, Korshunova GA, Lyamzaev KG, et al. Mitochondrial-targeted plastoquinone derivatives. Effect on senescence and acute age-related pathologies. *Curr Drug Targets*. 2011 Jun;12(6):800-26.
- Swerdlow RH, Burns JM, Khan SM. The Alzheimer's disease mitochondrial cascade hypothesis: progress and perspectives. *Biochim Biophys Acta*. 2014 Aug;1842(8):1219-31.
- Prince M, Bryce R, Albanese E, Wimo A, Ribeiro W, Ferri CP. The global prevalence of dementia: a systematic review and metaanalysis. *Alzheimers Dement*. 2013 Jan;9(1):63-75 e2.
- Smith MA, Perry G. The pathogenesis of Alzheimer disease: an alternative to the amyloid hypothesis. *J Neuropathol Exp Neurol*. 1997 Feb;56(2):217.
- Alzheimer's A. 2016 Alzheimer's disease facts and figures. *Alzheimers Dement*. 2016 Apr;12(4):459-509.
- Pagani L, Eckert A. Amyloid-Beta interaction with mitochondria. *Int J Alzheimers Dis*. 2011 Mar 15;2011:925050.
- Price DL, Sisodia SS, Borchelt DR. Alzheimer disease—when and why? *Nat Genet*. 1998 Aug;19(4):314-6.
- Calkins MJ, Manczak M, Mao P, Shirendeb U, Reddy PH. Impaired mitochondrial biogenesis, defective axonal transport of mitochondria, abnormal mitochondrial dynamics and synaptic degeneration in a mouse model of Alzheimer's disease. *Hum Mol Genet*. 2011 Dec 01;20(23):4515-29.
- Matic I, Strobbe D, Frison M, Campanella M. Controlled and Impaired Mitochondrial Quality in Neurons: Molecular Physiology and Prospective Pharmacology. *Pharmacol Res*. 2015 Sep;99:410-24.
- Yan MH, Wang X, Zhu X. Mitochondrial defects and oxidative stress in Alzheimer disease and Parkinson disease. *Free Radic Biol Med*. 2013 Sep;62:90-101.

- 15 Wang X, Su B, Siedlak SL, Moreira PI, Fujioka H, Wang Y, Casadesus G, Zhu X. Amyloid-beta overproduction causes abnormal mitochondrial dynamics via differential modulation of mitochondrial fission/fusion proteins. *Proc Natl Acad Sci U S A*. 2008 Dec 09;105(49):19318-23.
- 16 McManus MJ, Murphy MP, Franklin JL. The mitochondria-targeted antioxidant MitoQ prevents loss of spatial memory retention and early neuropathology in a transgenic mouse model of Alzheimer's disease. *J Neurosci*. 2011 Nov 02;31(44):15703-15.
- 17 Bose A, Beal MF. Mitochondrial dysfunction in Parkinson's disease. *J Neurochem*. 2016 Oct;139 Suppl 1:216-231.
- 18 Basso M, Giraud S, Corpillo D, Bergamasco B, Lopiano L, Fasano M. Proteome analysis of human substantia nigra in Parkinson's disease. *Proteomics*. 2004 Dec;4(12):3943-52.
- 19 Obeso JA, Rodriguez-Oroz MC, Goetz CG, Marin C, Kordower JH, Rodriguez M, Hirsch EC, Farrer M, Schapira AH, Halliday G. Missing pieces in the Parkinson's disease puzzle. *Nat Med*. 2010 Jun;16(6):653-61.
- 20 Jin H, Kanthasamy A, Ghosh A, Anantharam V, Kalyanaraman B, Kanthasamy AG. Mitochondria-targeted antioxidants for treatment of Parkinson's disease: preclinical and clinical outcomes. *Biochim Biophys Acta*. 2014 Aug;1842(8):1282-94.
- 21 Sandy MS, Langston JW, Smith MT, Di Monte DA. PCR analysis of platelet mtDNA: lack of specific changes in Parkinson's disease. *Mov Disord*. 1993 8(1):74-82.
- 22 Ramsay RR, Kowal AT, Johnson MK, Salach JI, Singer TP. The inhibition site of MPP<sup>+</sup>, the neurotoxic bioactivation product of 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine is near the Q-binding site of NADH dehydrogenase. *Arch Biochem Biophys*. 1987 Dec;259(2):645-9.
- 23 Schapira AH, Cooper JM, Dexter D, Clark JB, Jenner P, Marsden CD. Mitochondrial complex I deficiency in Parkinson's disease. *J Neurochem*. 1990 Mar;54(3):823-7.
- 24 Gash DM, Rutland K, Hudson NL, Sullivan PG, Bing G, Cass WA, Pandya JD, Liu M, Choi DY, Hunter RL, et al. Trichloroethylene: Parkinsonism and complex 1 mitochondrial neurotoxicity. *Ann Neurol*. 2008 Feb;63(2):184-92.
- 25 Panov A, Dikalov S, Shalbuyeva N, Taylor G, Sherer T, Greenamyre JT. Rotenone model of Parkinson disease: multiple brain mitochondria dysfunctions after short term systemic rotenone intoxication. *J Biol Chem*. 2005 Dec 23;280(51):42026-35.
- 26 Borland MK, Trimmer PA, Rubinstein JD, Keeney PM, Mohanakumar K, Liu L, Bennett JP, Jr. Chronic, low-dose rotenone reproduces Lewy neurites found in early stages of Parkinson's disease, reduces mitochondrial movement and slowly kills differentiated SH-SY5Y neural cells. *Mol Neurodegener*. 2008 Dec 29;3:21.
- 27 Perier C, Bove J, Dehay B, Jackson-Lewis V, Rabinovitch PS, Przedborski S, Vila M. Apoptosis-inducing factor deficiency sensitizes dopaminergic neurons to parkinsonian neurotoxins. *Ann Neurol*. 2010 Aug;68(2):184-92.
- 28 Thomas B, Beal MF. Parkinson's disease. *Hum Mol Genet*. 2007 Oct 15;16 Spec No. 2:R183-94.
- 29 Liu G, Zhang C, Yin J, Li X, Cheng F, Li Y, Yang H, Ueda K, Chan P, Yu S. alpha-Synuclein is differentially expressed in mitochondria from different rat brain regions and dose-dependently down-regulates complex I activity. *Neurosci Lett*. 2009 May 01;454(3):187-92.
- 30 Pickrell AM, Youle RJ. The roles of PINK1, parkin, and mitochondrial fidelity in Parkinson's disease. *Neuron*. 2015 Jan 21;85(2):257-73.
- 31 Kazlauskaitė A, Muqit MM. PINK1 and Parkin - mitochondrial interplay between phosphorylation and ubiquitylation in Parkinson's disease. *FEBS J*. 2015 Jan;282(2):215-23.
- 32 Eiyama A, Okamoto K. PINK1/Parkin-mediated mitophagy in mammalian cells. *Curr Opin Cell Biol*. 2015 Apr;33:95-101.
- 33 Krebiehl G, Ruckerbauer S, Burbulla LF, Kieper N, Maurer B, Waak J, Wolburg H, Gizatullina Z, Gellerich FN, Voitalla D, et al. Reduced basal autophagy and impaired mitochondrial dynamics due to loss of Parkinson's disease-associated protein DJ-1. *PLoS One*. 2010 Feb 23;5(2):e9367.
- 34 Parkinson Study Group QE1, Beal MF, Oakes D, Shoulson I, Henchcliffe C, Galpern WR, Haas R, Juncos JL, Nutt JG, Voss TS, et al. A randomized clinical trial of high-dosage coenzyme Q10 in early Parkinson disease: no evidence of benefit. *JAMA Neurol*. 2014 May;71(5):543-52.
- 35 Snow BJ, Rolfe FL, Lockhart MM, Frampton CM, O'Sullivan JD, Fung V, Smith RA, Murphy MP, Taylor KM, Protect Study G. A double-blind, placebo-controlled study to assess the mitochondria-targeted antioxidant MitoQ as a disease-modifying therapy in Parkinson's disease. *Mov Disord*. 2010 Aug 15;25(11):1670-4.
- 36 Ross MF, Kelso GF, Blaikie FH, James AM, Cocheme HM, Filipovska A, Da Ros T, Hurd TR, Smith RA, Murphy MP. Lipophilic triphenylphosphonium cations as tools in mitochondrial bioenergetics and free radical biology. *Biochemistry (Mosc)*. 2005 Feb;70(2):222-30.
- 37 Smith RA, Porteous CM, Gane AM, Murphy MP. Delivery of bioactive molecules to mitochondria in vivo. *Proc Natl Acad Sci U S A*. 2003 Apr 29;100(9):5407-12.
- 38 Kelso GF, Porteous CM, Coulter CV, Hughes G, Porteous WK, Ledgerwood EC, Smith RA, Murphy MP. Selective targeting of a redox-active ubiquinone to mitochondria within cells: antioxidant and antiapoptotic properties. *J Biol Chem*. 2001 Feb 16;276(7):4588-96.
- 39 Oyewole AO, Birch-Machin MA. Mitochondria-targeted antioxidants. *FASEB J*. 2015 Dec;29(12):4766-71.
- 40 Ramis MR, Esteban S, Miralles A, Tan DX, Reiter RJ.

- Protective Effects of Melatonin and Mitochondria-targeted Antioxidants Against Oxidative Stress: A Review. *Curr Med Chem*. 2015 22(22):2690-711.
- 41 Smith RA, Murphy MP. Mitochondria-targeted antioxidants as therapies. *Discov Med*. 2011 Feb;11(57):106-14.
- 42 Magwere T, West M, Riyahi K, Murphy MP, Smith RA, Partridge L. The effects of exogenous antioxidants on lifespan and oxidative stress resistance in *Drosophila melanogaster*. *Mech Ageing Dev*. 2006 Apr;127(4):356-70.
- 43 James AM, Sharpley MS, Manas AR, Frerman FE, Hirst J, Smith RA, Murphy MP. Interaction of the mitochondria-targeted antioxidant MitoQ with phospholipid bilayers and ubiquinone oxidoreductases. *J Biol Chem*. 2007 May 18;282(20):14708-18.
- 44 Doughan AK, Dikalov SI. Mitochondrial redox cycling of mitoquinone leads to superoxide production and cellular apoptosis. *Antioxid Redox Signal*. 2007 Nov;9(11):1825-36.
- 45 Liu X, Murphy MP, Xing W, Wu H, Zhang R, Sun H. Mitochondria-targeted antioxidant MitoQ reduced renal damage caused by ischemia-reperfusion injury in rodent kidneys: Longitudinal observations of T2-weighted imaging and dynamic contrast-enhanced MRI. *Magn Reson Med*. 2017 Jun 12. doi: 10.1002/mrm.26772.
- 46 Escribano-Lopez I, Diaz-Morales N, Rovira-Llopis S, de Maranon AM, Orden S, Alvarez A, Banuls C, Rocha M, Murphy MP, Hernandez-Mijares A, et al. The mitochondria-targeted antioxidant MitoQ modulates oxidative stress, inflammation and leukocyte-endothelium interactions in leukocytes isolated from type 2 diabetic patients. *Redox Biol*. 2016 Dec;10:200-205.
- 47 Graham D, Huynh NN, Hamilton CA, Beattie E, Smith RA, Cocheme HM, Murphy MP, Dominiczak AF. Mitochondria-targeted antioxidant MitoQ10 improves endothelial function and attenuates cardiac hypertrophy. *Hypertension*. 2009 Aug;54(2):322-8.
- 48 McManus MJ, Murphy MP, Franklin JL. Mitochondria-derived reactive oxygen species mediate caspase-dependent and -independent neuronal deaths. *Mol Cell Neurosci*. 2014 Nov;63:13-23.
- 49 Ng LF, Gruber J, Cheah IK, Goo CK, Cheong WF, Shui G, Sit KP, Wenk MR, Halliwell B. The mitochondria-targeted antioxidant MitoQ extends lifespan and improves healthspan of a transgenic *Caenorhabditis elegans* model of Alzheimer disease. *Free Radic Biol Med*. 2014 Jun;71:390-401.
- 50 Solesio ME, Prime TA, Logan A, Murphy MP, Del Mar Arroyo-Jimenez M, Jordan J, Galindo MF. The mitochondria-targeted anti-oxidant MitoQ reduces aspects of mitochondrial fission in the 6-OHDA cell model of Parkinson's disease. *Biochim Biophys Acta*. 2013 Jan;1832(1):174-82.
- 51 Ghosh A, Chandran K, Kalivendi SV, Joseph J, Antholine WE, Hillard CJ, Kanthasamy A, Kanthasamy A, Kalyanaraman B. Neuroprotection by a mitochondria-targeted drug in a Parkinson's disease model. *Free Radic Biol Med*. 2010 Dec 01;49(11):1674-84.
- 52 Rodriguez-Cuenca S, Cocheme HM, Logan A, Abakumova I, Prime TA, Rose C, Vidal-Puig A, Smith AC, Rubinsztein DC, Fearnley IM, et al. Consequences of long-term oral administration of the mitochondria-targeted antioxidant MitoQ to wild-type mice. *Free Radic Biol Med*. 2010 Jan 01;48(1):161-72.
- 53 Gane EJ, Weilert F, Orr DW, Keogh GF, Gibson M, Lockhart MM, Frampton CM, Taylor KM, Smith RA, Murphy MP. The mitochondria-targeted antioxidant mitoquinone decreases liver damage in a phase II study of hepatitis C patients. *Liver Int*. 2010 Aug;30(7):1019-26.
- 54 Kim HK, Han J. Mitochondria-Targeted Antioxidants for the Treatment of Cardiovascular Disorders. *Adv Exp Med Biol*. 2017 982:621-646.
- 55 Park SY, Kwon OS, Andtbacka RHI, Hynstrom JR, Reese V, Murphy MP, Richardson RS. Age-related endothelial dysfunction in human skeletal muscle feed arteries: the role of free radicals derived from mitochondria in the vasculature. *Acta Physiol (Oxf)*. 2017 May 11. doi: 10.1111/apha.12893.