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The Parkinson Disease Mitochondrial Hypothesis: Where Are We at?

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Abstract

Parkinson's disease is a common, adult-onset neurodegenerative disorder whose pathogenesis is still under intense investigation. Substantial evidence from postmortem human brain tissue, genetic- and toxin-induced animal and cellular models indicates that mitochondrial dysfunction plays a central role in the pathophysiology of the disease. This review discusses our current understanding of Parkinson's disease–related mitochondrial dysfunction, including bioenergetic defects, mitochondrial DNA alterations, altered mitochondrial dynamics, activation of mitochondrial-dependent programmed cell death, and perturbations in mitochondrial tethering to the endoplasmic reticulum. Whether a primary or secondary event, mitochondrial dysfunction holds promise as a potential therapeutic target to halt the progression of neurodegeneration in Parkinson's disease.

Keywords

complex I, apoptosis, fusion/fission, mtDNA, mitochondria-associated endoplasmic reticulum membranes

Introduction

Parkinson's disease (PD) is the second most common neurodegenerative disease after Alzheimer's dementia, affecting about 2% of the population older than 60 years. PD patients typically experience difficulties with slowness of movements (bradykinesia), stiffness of the muscles (rigidity), tremor, and balance disturbances. In addition to this, PD is known to have profound effects on both cognition and mood. So far, only symptomatic treatments are available, and although they are very helpful, particularly in the early stages of the disease, the therapeutic benefit during long-term treatment gradually declines and most patients develop motor and psychiatric side effects, which may be as debilitating as the disease itself. To date, there are no disease-modifying therapies that directly target the underlying disease mechanisms or halt progression of the disease. Elucidation of the molecular mechanisms underlying the pathogenesis of PD may represent the most fundamental basis for the development of disease-modifying therapies. The specific pathophysiology responsible for all of the symptoms observed in PD is still under investigation, but the severe depletion of dopamine (DA) within the striatum resulting from the extensive loss of DA-containing neurons in the substantia nigra pars compacta (SNpc) is considered to be the predominant histological feature. Why specific subsets of neurons are particularly vulnerable to degeneration is one of the central unresolved mysteries of neurodegenerative disease research, and the driving force behind the intense investigation currently being carried out to elucidate the etiopathogenesis of PD. To this end, various hypotheses regarding the pathogenesis of the disease suggest that the mechanisms underlying dopaminergic cell death might be the consequence of mitochondrial dysfunction, alteration of protein degradation pathways, misfolding and aggregation of proteins such as α-synuclein, and/or the presence of neuroinflammation. In this review, we aim to summarize the current knowledge of mitochondrial biology and to demonstrate how recent evidence concerning mitochondrial dysfunction has shed light on the role of mitochondria in PD.

Mitochondria were first identified 110 years ago in eukaryotic cells, and described as a collection of free-floating individual vesicles existing in hundreds of copies, remaining freely in the cytosol. The main hypothesis of the evolutionary origin of mitochondria suggests that they originated from a symbiotic relationship between eukaryotic cells and primitive bacteria capable of oxidative

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phosphorylation. This hypothesis was supported by the discovery of DNA in mitochondria and by the fact that mitochondria were found to have their own machinery for protein synthesis. The primary function of mitochondria is the generation of cellular energy in the form of adenosine 5′-triphosphate (ATP) by oxidative phosphorylation. As such, mitochondria are usually considered the "powerhouses of the cell." However, mitochondrial biological function extends well beyond just the production of energy; they are highly dynamic organelles with complex structural features that have important cellular functions such as the metabolism of amino acids and lipids, as well as housing intermediate metabolic pathways such as pyruvate oxidation, the Krebs cycle, the regulation of calcium homeostasis, free radical scavenging or the control of programmed cell death (PCD).

Cells contain many mitochondria, each of which carries several copies of a small circular mitochondrial genome in the matrix. While it was thought that the mitochondrion was a rigid organelle, it is now clear that mitochondria undergo constant morphological changes due to repetitive cycles of fusion (the combination of two mitochondria into a single organelle) and fission (the separation of long, tubular mitochondria into two or more smaller parts), resulting in a wide range of mitochondrial morphologies. This phenomenon is regulated by a delicate balance between these two opposing processes; in turn, the balance influences most mitochondrial functions and allows mitochondrial integrity, bioenergetic function, mitochondrial turnover, and protection of mtDNA to be maintained.

Do Alterations to Mitochondrial Respiration Participate in PD Neurodegeneration?

Over the past 25 years, publications in the field of PD-related mitochondrial dysfunction have increased exponentially in number and many studies have demonstrated that alterations in mitochondrial function are central to the pathogenesis of PD.

Several lines of evidence sustain a link between sporadic PD and a dysfunctional respiratory chain, in particular a deficit in complex I activity. In 1983, Langston and others observed that accidental exposure of drug abusers to 1-methyl-4-phenyl-1,2,3,4-tetrahydropyridine (MPTP), an inhibitor of complex I (NADH/ubiquinone oxidoreductase) of the mitochondrial electron transport chain, resulted in an acute and irreversible parkinsonian syndrome that was almost indistinguishable from PD. In 2000, the group led by Greenamyre reported that the chronic, systemic inhibition of complex I by the pesticide rotenone, causes highly selective nigrostriatal dopaminergic degeneration (Betarbet and others 2000). Finally, a biochemical link between complex I inhibition and sporadic PD was established when several groups reported reduced complex I activity in the brains of patients with PD (Hattori and others 1991; Schapira and others 1990). It was also demonstrated that catalytic subunits of complex I derived from PD frontal cortex mitochondria are oxidatively damaged, correlating with complex I misassembly and dysfunction (Keeney and others 2006). However, diminished activity in complex I has not only been reported in the postmortem substantia nigra but also seems to extend to multiple neuronal and non-neuronal regions, including the cortex, skeletal muscles, fibroblasts and platelets of PD patients (Parker and others 1989; Schapira and others 1990). Thus, the relevance of complex I inhibition to disease pathogenesis remains uncertain and several key questions remain to be explained: (a) How does a systemic defect cause selective degeneration of the dopamine neurons of the substantia nigra pars compacta? (b) Knowing that complex I activity should be reduced by more than 50% to cause significant ATP depletion in non-synaptic brain mitochondria (Davey and Clark 1996), why is it that a reduction of only 25% to 30% of complex I activity in PD patients (Schapira and others 1990) might play a major role in energy failure in PD-related dopaminergic neurodegeneration? (c) Does a deficit in complex I represent a primary or secondary event in the pathogenesis of PD?

Our own view is that the complex I defects are likely to be critical in the pathogenesis of dopaminergic neuronal loss, probably in combination with other defects (Fig. 1). Indeed, we showed that structural alterations to complex I linked to a deficiency in apoptosis-inducing factor (AIF) do not cause dopaminergic neurodegeneration *per se*, but increase the susceptibility of dopaminergic neurons to exogenous parkinsonian neurotoxins (Perier and others 2010). However, it still remains to be elucidated if the majority of PD patients or only a portion of them present a complex I deficiency.

Nonetheless, even if energy failure might not be responsible for DA cell death, it is now clear that mitochondrial dysfunction induces the chronic production of reactive oxygen species (ROS) and is instrumental in the demise of DA neurons. ROS include the superoxide anion ($O_2^{\cdot -}$), hydrogen peroxide (H_2O_2), and the hydroxyl radical ($OH^{\cdot}$). H_2O_2 and $OH^{\cdot}$ are potent oxidants that extract electrons from other molecules such as DNA, proteins and lipids, thereby altering their properties. Complex I is known to be the major source of superoxide production in the electron transport chain in presence or absence of toxins. Oxidative damage to proteins, lipids and DNA has been observed in postmortem brain samples from PD patients (Dauer and Przedborski 2003), while the PD-linked protein DJ-1, mutations of which cause an autosomal recessive form of PD (Bonifati and others 2003), has been identified as a

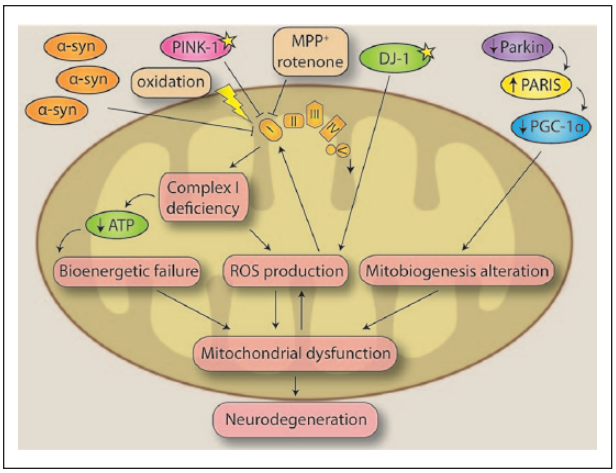

Figure 1. Alterations to mitochondrial respiration in Parkinson's disease (PD). Alterations to several mitochondria-based mechanisms may produce energy failure, mitochondrial dysfunction, and potentially lead to neuronal death. The drawing schematizes how a deficit in oxidative phosphorylation induced by (a) mitochondrial neurotoxins (MPP^+ or rotenone), (b) overexpression of α-synuclein, or (c) PINK1 mutations, leads to complex I inhibition, ATP depletion, or bioenergetic failure. Increased reactive oxygen species (ROS) production due to the leakage of electrons through the respiratory chain feeds a vicious cycle, since oxygen radicals act on complex I leading to its deficiency. PD-linked DJ-1 mutations result in increased mitochondrial ROS production. Accumulation of PARIS, a Parkin-interacting substrate, may be one of the mechanisms that can explain the PGC-1α down-regulation observed in PD, which probably induces a decrease in mitobiogenesis.

mitochondrial peroxiredoxin-like peroxidase, able to scavenge mitochondrial ROS (Andres-Mateos and others 2007) (Fig. 1). Moreover, supporting an instrumental role for mitochondrial-derived ROS in PD-related neurodegeneration, transgenic mice overexpressing mitochondrially targeted catalase exhibited an attenuation of MPTP-induced mitochondrial ROS and dopaminergic cell death (Perier and others 2010).

More recently, increasing evidence has demonstrated that mitochondrial dysfunction may be the consequence of alterations to mitobiogenesis through the deregulation of transcription factors, thus perturbing cellular bioenergetics. In particular, proliferator-activated receptor g coactivator-1α (PGC-1α), a coactivator of several transcription factors, and a key regulator of mitochondrial biogenesis, was found to be decreased in PD (Zheng and others 2010). Evidence also points to a role for PGC-1α in disease pathogenesis. For example (a) studies employing PGC-1α knockout mice revealed an increased sensitivity of dopaminergic cells to MPTP (St-Pierre and others 2006), (b) transgenic overexpression of PGC-1α is neuroprotective against MPTP-induced neuroxicity (Mudò and others 2012), and (c) pharmacological activation of PGC-1α by resveratrol produced a similar degree of neuroprotection to dopaminergic neurons as that observed with the transgenic overexpression of PGC-1α (Mudò and others 2012). These observations suggest that PGC-1α could be a promising therapeutic target for PD treatment (Fig. 1), although this remains a matter of debate as PGC-1α overexpression in the rat nigrostriatal system induced an unexpected degeneration of dopaminergic neurons (Ciron and others 2012). Nonetheless, it is worth noting that PGC-1α deficiency is associated with increased α-synuclein oligomerization, while, inversely, the overexpression of PGC-1α reduced α-synuclein oligomerization and rescued cells from α-synuclein-mediated toxicity (Eschbach and others 2015).

Mitochondrial DNA: The Achilles Heel of PD?

The mitochondrial genome consists of a multicopy, double-stranded, circular molecule (16.6 kb in humans), which encodes 13 essential polypeptides of the respiratory chain and the necessary RNA machinery for their translation within the organelle (2 rRNAs and 22 tRNAs). Mitochondrial genetics differ from Mendelian genetics in almost every aspect: maternal inheritance, presence of many copies of the genome within a single cell, and the mechanisms underlying replication. Mitochondrial DNA (mtDNA) mutations accumulate with age and it is thought that once the pathogenic threshold in a previously unaffected tissue is surpassed, the phenotype can change; this may explain the age-related, and even tissue-related, variability of clinical features. The threshold for disease is lower in tissues that are highly dependent on oxidative metabolism, such as brain, heart and skeletal muscle, rendering these tissues especially vulnerable to the effects of pathogenic mutations in mtDNA.

To date no PD-associated genetic mutations in mtDNA have been identified. However, it seems that haplogroups J, K are protective against the development of PD within European populations and that super haplogroup HV is associated with an increased risk of developing PD with advancing age (Hudson and others 2013). These data interestingly suggest that subtle changes within the mitochondrial genetic makeup may play a role in the development of PD. Remarkably, mtDNA deletions have been observed in individual dopaminergic neurons microdissected from the substantia nigra of postmortem human brains from idiopathic PD patients (Bender and others 2006). Although their effects on bioenergetic function are difficult to estimate, high levels of mtDNA deletions in these neurons were associated with a decreased histochemical activity of cytochrome c oxidase, one of the key enzymes of the

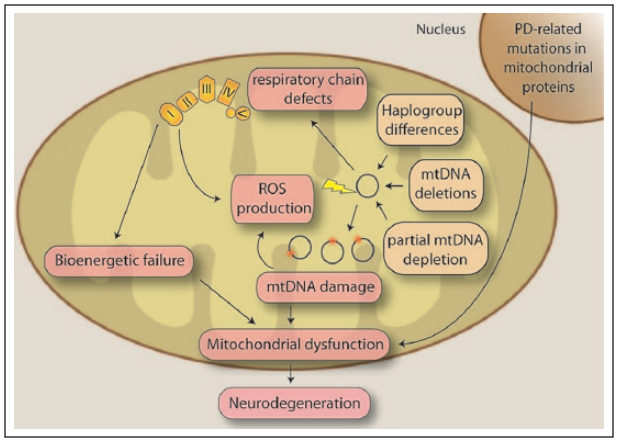

Figure 2. Mitochondrial DNA alterations and Parkinson's disease. Several mitochondrial DNA (mtDNA) alterations together with mtDNA genetic differences account for mitochondrial DNA damage in a given mitochondrion. These alterations can increase reactive oxygen species (ROS) production, which in turn can damage mtDNA. Under normal conditions, all mtDNAs within a cell are identical (homoplasmy). Under pathological conditions linked to pathogenic mtDNA mutations, cells can harbor both normal and mutant mtDNA (heteroplasmy). In the latter case, a minimum number of mutated mtDNAs is required to cause mitochondrial dysfunction and clinical signs (threshold effect). Apart from mtDNA alterations, mutations linked to PD in nuclear-encoded mitochondrial proteins, such as PINK1, Parkin, α-synuclein, DJ-1, and leucine-rich-repeat kinase 2 (LRRK2) also lead to mitochondrial dysfunction.

respiratory chain, suggesting that accumulation of mtDNA deletions over a certain threshold in SNpc dopaminergic neurons may cause mitochondrial functional defects associated with PD (Bender and others 2006) (Fig. 2). However, because of the circumstantial nature of data obtained from postmortem samples, it cannot be excluded that mtDNA deletions represent a secondary rather than a primary event in the pathogenesis of PD. Attempts were made to stress the primary role of mtDNA alterations by using mutant mice possessing an amino acid substitution (D257A) in the exonuclease domain II of POLG that ablates its proofreading activity; this induced an accumulation of mtDNA deletions (Kujoth and others 2005). We and others showed that despite high levels of mtDNA deletions, SNpc dopaminergic neurons from these animals did not exhibit gross mitochondrial dysfunction or degeneration (Dai and others 2014; Perier and others 2013). These results suggest that primary accumulation of high levels of mtDNA deletions below a certain threshold is not sufficient *per se* to kill substantia nigra pars compacta dopaminergic neurons. In contrast, partial depletion of mtDNA in mice, either by a conditional disruption in dopaminergic neurons of mitochondrial transcription factor A (TFAM), which regulates mitochondrial DNA transcription (Ekstrand and others 2007), or by expression of a mitochondria-targeted restriction enzyme (PstI), which induces double-strand breaks in mtDNA (Pickrell and others 2011), leads to respiratory chain defects and slowly progressing, levodopa-responsive motor deficits associated with nigrostriatal denervation. These findings clearly demonstrate that primary respiratory chain deficiency in DA neurons can lead to a progressive parkinsonian phenotype in mice, stressing the importance of mtDNA integrity in PD.

Finally, apart from mtDNA mutations/deletions and depletions, the presence of mtDNA damage specifically in the SNpc in the form of abasic sites able to block the polymerase during replication and transcription, which can result from and can cause oxidative stress and mitochondrial impairment, was recently described in postmortem tissue of PD patients (Sanders and others 2014).

Disruption of Key Mitochondrial Processes

Mitochondria, a Dynamic Organelle

Mitochondria can change their shape, size and inner membrane structure in a dynamic fashion, and the mitochondria of a single cell do not function in isolation, but form a complex reticulum whose morphology undergoes continuous cycles of fusion and fission (Twig and Shirihai 2011). These opposing processes determine the architecture of the entire mitochondrial population for which a strict control of such events is essential for maintaining the metabolic function of mitochondria. Indeed, respiration, calcium buffering, and apoptosis are influenced by mitochondrial dynamics. Dysfunction of this system, whether by toxic insult or by genetic defect, may lead to cell death and induce neurodegeneration. At the molecular level, mitochondrial fusion is mediated by a group of GTPases: mitofusins (MFN1 and MNF2) at the outer membrane, and OPA1 in the inner membrane. In addition to its role in fusion, OPA1 is also responsible for maintaining the structure of the cristae (Frezza and others 2006). Mitochondrial fission requires the recruitment of a dynamin-related protein (DRP1) from the cytosol which is thought to assemble into rings and spirals that encircle and constrict the mitochondrial tubule during fission (Detmer and Chan 2007) (Fig. 3A). Studies of mitochondrial morphology in brain tissue from the SNpc of PD patients have been limited for various reasons. First, autopsy tissue from patients is available, but ultrastructural details are often barely preserved. Second, brain biopsy samples, which are more difficult to obtain, are often from patients in later stages of the disease at a time where pathological changes may no longer reflect the pathogenic process. Nonetheless, various parkinsonian-related neurotoxic molecules cause mitochondrial

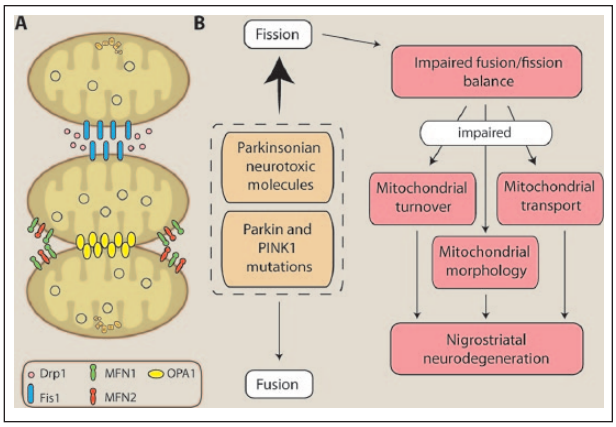

Figure 3. Mitochondrial dynamics. (A) Molecules implicated in mitochondrial fusion and fission, which control mitochondrial number and size. For fission to take place, dynamin-related protein 1 (Drp1) needs to be recruited to the outer mitochondrial membrane together with the mitochondrial fission-1 protein (Fis1). Mitofusins 1 and 2 (MFN1, MFN2) are needed for the tethering of the outer mitochondrial membrane, while optic atrophy 1 (OPA1) is responsible for inner mitochondrial membrane fusion. (B) An imbalance in mitochondrial dynamics, either due to environmental neurotoxins or due to Parkinson's disease–linked mutations of Parkin and PINK1, affects mitochondrial morphology, mitochondrial turnover and mitochondrial transport, eventually giving rise to nigrostriatal neurodegeneration.

fission in vitro (Santos and Cardoso 2012). Only recently, several PD related genes were shown to play critical roles in maintaining normal mitochondrial morphology and function. Indeed, the PD-related proteins PINK1 and Parkin appear to control mitochondrial morphology by regulating mitochondrial fusion/fission events (for review, see Scarffe and others 2014). It is now clear that a fine a balance between fusion and fission is crucial not only to mitochondrial morphology, but also to cell viability and synaptic function (Rappold and others 2014). In addition to morphological changes, evidence implicates the Parkin/PINK1 pathway in various mitochondrial functions, including mitophagy, transport, and biogenesis (Scarffe and others 2014). Mitochondrial transport and subcellular distribution are also of importance for neuronal function. In neurons, mitochondria are particularly abundant at synapses and concentrate in subcellular regions with high metabolic requirements, for example close to active growth cones in neurons (Chang and others 2006). Drp1 is critical for targeting mitochondria to the nerve terminal, and a disruption in mitochondrial fission can contribute to the preferential death of nigrostriatal DA neurons (Berthet and others 2014). Taken together, these experimental results stress the importance of defects in mitochondrial dynamics in the pathogenesis of PD (Fig. 3B).

Mitochondria: Beyond Life, the Conductor of Cell Death

Although substantial evidences show that mitochondrial dysfunction plays a crucial role in the pathogenesis of PD, the exact mechanism underlying cell death in PD is still under investigation. In response to various stressors, cells possess "programmed" physiological processes, named programmed cell death (PCD), which is activated to cause their destruction. The term PCD is often used interchangeably with apoptosis; however, it is now well established that other types of PCD, such as programmed necrosis, also occur in the same types of cells. The different forms of PCD can be activated by the same stimulus, depending on its nature (acute vs. chronic), its duration, and the bioenergetic state of the cell, thus adding a level of complexity to the disease's pathogenesis.

The most common form of PCD is apoptosis, a molecular process that occurs naturally during development and morphogenesis. Apoptosis is composed of two main pathways, an extrinsic one that can bypass the mitochondria, and an intrinsic one that requires mitochondrial involvement. Whereas several components of the mitochondrial apoptotic pathway have been implicated in the pathogenesis of PD, participation of the extrinsic pathway in PD has not been shown in a consistent manner. As reviewed elsewhere (Perier and Vila 2012), we and others showed that inhibition of complex I blocks the flow of electrons along the mitochondrial electron transport chain, resulting in an increased production of ROS. Mitochondrial ROS then damage different cellular elements, such as lipids, proteins, and DNA. Moreover, complex I–dependent disruption of OPA1 oligomeric complexes that normally keep mitochondrial cristae junctions tight, allows the mobilization of cytochrome c from cristae to the intermembrane space, thereby lowering the threshold for activation of mitochondria-dependent apoptosis by cell death agonists in compromised neurons (Ramonet and others 2013). In parallel with this, DNA damage activates pro-apoptotic pathways, inducing mitochondrial translocation of Bax. Once localized to the mitochondrial outer membrane, Bax induces the release of cytochrome c into the cytosol, with ensuing caspase activation and cell death (Perier and others 2005; Perier and others 2007) (Fig. 4). Supporting the relevance of this scenario to the SNpc dopaminergic neurodegeneration that occurs in PD, several elements of this molecular cascade have been demonstrated in postmortem human brain samples from PD patients, including complex I deficiency, ROS production, oxidative damage to lipids, proteins and DNA, JNK activation, Bax activation and activation of caspase-3 and -9 (for review, see Perier and others 2012).

PD gene products have also been linked to apoptosis. Overexpressing PD-linked LRRK2 mutations induce

apoptosis in vitro (Iaccarino and others 2007). On the other hand, wild-type DJ-1 protects cells from apoptosis (Venderova and Park 2012), whereas DJ-1 deficiency increases oxidative stress-induced apoptotic cell death (Martinat and others 2004). Overexpression of wild-type PINK1, but not of PD-associated PINK1 mutants, is able to attenuate cytochrome c release (Wang and others 2007). Parkin prevents cytochrome c release (Darios and others 2003) and apoptosis by the inhibition of Bax translocation (Charan and others 2014), while the overexpression of α-synuclein is associated with complex I inhibition (Loeb and others 2010) (Fig. 4). A recent study showed that human α-synuclein in yeast cells also triggers mitochondria-nuclear translocation of EndoG and EndoG-mediated DNA degradation (Büttner and others 2013), indicating that α-synuclein toxicity may activate apoptosis as well as caspase-independent cell death.

Among the caspase-independent programmed cell death scenarios, necrosis, which has traditionally been considered as passive cell death, is now considered a genetically programmed pathway with necrotic morphology. Programmed necrosis (or necroptosis) is associated with a rapid bioenergetic failure, ROS formation, and loss of plasma membrane integrity. Relatively few studies have been performed to study the involvement of programmed necrosis in the pathogenesis of PD, as for a long time, studying PCD in PD was synonymous with searching for apoptotic cells in post mortem tissue and in models of the disease. Nonetheless, recent evidence suggests its implication, and in particular the involvement of excessive activation of poly(ADP-ribose) polymerase-1 (PARP-1), which gives rise to a caspase-independent form of PCD, termed "parthanatos.". PARP-1 activation is an early biochemical cell death event that is to a large extent mediated by the accumulation of poly (ADP-ribose) (PAR) and nuclear translocation of AIF from the mitochondria. The activation of PARP-1 contributes to MPTP-induced dopaminergic neurodegeneration (Wang and others 2009), as demonstrated by PARP−/− mice, which are resistant to MPTP intoxication (Mandir and others 1999). This finding led the team of Dawson to propose that excessive activation of PARP-1 leads to parthanatos, in which PAR polymer appears to be a pro-death signaling molecule that acts as a nuclear-mitochondrial signal to release AIF from the mitochondria in PARP-1-dependent cell death (Andrabi and others 2008) (Fig. 4). Although PAR polymer involvement was not demonstrated in the context of the pathogenesis of PD, in vitro experiments have shown that AIF can be released from the mitochondria, along with cytochrome c, following complex I inhibition with MPP^+, the active metabolite of MPTP (Chu and others 2005). Furthermore, small interfering RNA-mediated knockdown of AIF in dopaminergic cell lines is able to delay MPP^+-induced cell death (Chu and others 2005). However, whether AIF plays an actual pro-cell death role in experimental in vivo models of PD remains to be demonstrated, as mutant mice partially deficient for AIF are not only not protected against MPTP intoxication but are also much more sensitive to the deleterious effects of this parkinsonian neurotoxin (Perier and others 2010).

Alterations to Mitophagy, an Emerging Problem for PD

Autophagy is a process whereby the cytoplasmic contents of a cell are sequestered within double membrane vacuoles, called autophagosomes (AP), and subsequently delivered to the lysosome for degradation. There are at least two ways in which alterations of autophagic pathways may participate in the demise of dopaminergic neurons. First, when the autophagic processes are insufficient to remove the accumulation of protein aggregates, such as α-synuclein. Second, the up-regulation of autophagy is considered to be a direct contributor to cell death. Increased numbers of AP have been observed in cultured cells exposed to parkinsonian neurotoxins such as MPP^+, rotenone and 6-OHDA (Dehay and others 2013), and in postmortem PD brain samples (Anglade 1997). However, controversy exists as to whether autophagy promotes or prevents cell death. If autophagy removes damaged mitochondria that would otherwise activate caspases and apoptosis, then autophagy should be protective. However, excessive and dysregulated autophagy may promote cell death, since enzymes leaking from lysosomes, such as cathepsins, can initiate mitochondrial permeabilization, caspase activation and apoptosis. In 2005, the term *mitophagy* for the selective autophagy of mitochondria was proposed (Lemasters 2005), and in recent years the link between mitophagy and PD has emerged as a new pathogenic hypothesis in PD. As reviewed elsewhere (Vives-Bauza and Przedborski 2011; Youle and Narendra 2011), the loss of mitochondrial membrane potential, which often correlates with dysfunctional mitochondria, seems to be a common signal for the mitochondria to be cleared via mitophagy. In mammalian cells, Parkin is recruited to depolarized mitochondria, which are subsequently eliminated by autophagy. Parkin translocation to mitochondria relies on PINK1 expression, suggesting that PINK1 is a key signaling molecule in mitophagy. Loss of DJ-1 leads to impaired autophagy, the accumulation of dysfunctional mitochondria (Krebiehl and others 2010), and a fragmented mitochondrial phenotype (Irrcher and others 2010). Interestingly, PINK1 and Parkin can rescue the mitochondrial fragmentation induced by the loss of DJ-1 (Irrcher and others 2010). These findings suggest that the different PD-related proteins linked to the mitochondria contribute to the pathogenesis of the disease at the intersection of mitochondrial dysfunction and autophagy (Fig. 4). However, to date, questions remain to be addressed: the majority of the

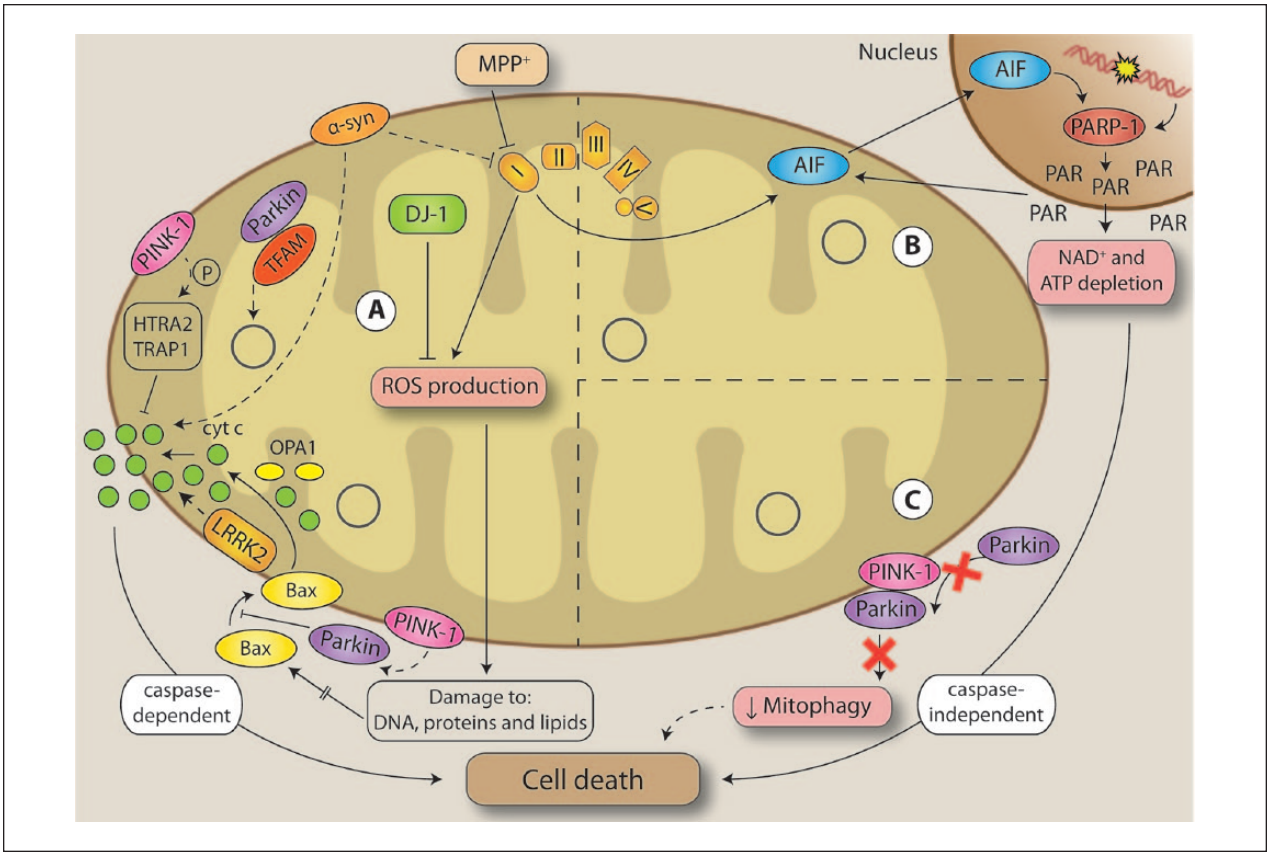

Figure 4. Activation of mitochondrial-dependent programmed cell death pathways in Parkinson's disease (PD). (A) Apoptosis can be activated following complex I inhibition, which gives rise to reactive oxygen species (ROS) production and the activation of intrinsic pro-apoptotic pathways (broken arrow; for more details see Perier and others 2012), which facilitate mitochondrial Bax translocation and release of cytochrome c (cyt c) into the cytosol. Mitochondrial PINK1 can attenuate cytochrome c release and apoptotic cell death by phosphorylating HTRA2 (high temperature requirement A2) or TRAP1 (tumor necrosis factor receptor-associated protein 1), two of its putative substrates. Mitochondrial Parkin, in turn, can prevent mitochondrial swelling, cytochrome c release, and apoptotic cell death. In addition, Parkin also regulates mitochondrial biogenesis through its interaction with the mitochondrial transcription factor A (TFAM). α-Synuclein (α-syn) has been shown to directly interact with mitochondria, where it can have a deleterious role by inhibiting complex I, increasing the production of ROS, and promoting the release of cytochrome c. LRRK2 (leucine-rich-repeat kinase 2) is partly localized to the mitochondria, where it can induce the release of cyt c and subsequent apoptotic cell death. (B) Necrosis can simultaneously occur with apoptosis. PARP-1 is activated in response to ROS-induced DNA damage, which results in excessive production of PAR polymers, a major product of PARP-1 activation, and NAD+ and ATP depletion. Such activation acts as a pro-death signal for the mitochondrion-to-nucleus translocation of AIF, which causes DNA fragmentation and programmed cell death. Mitochondrial outer membrane permeabilization represents the point-of-no-return in the mitochondrial apoptotic pathway, since once released into the cytosol, mitochondrial factors can initiate cell death in a caspase-dependent or a caspase-independent manner. (C) Mitophagy, the selective autophagic degradation of dysfunctional mitochondria, can be disrupted by PD-causing mutations in PINK1 and Parkin, thus resulting in the accumulation of undegraded, dysfunctional mitochondria that can cause cell dysfunction and ultimately cell death. Dashed arrows show unresolved pathways. Broken arrow represents described pathways.

studies performed to understand the mitophagic function of Parkin were performed in Parkin-overexpressing cells; at endogenous levels, Parkin fails to mediate mitophagy in human primary fibroblasts and in induced pluripotent stem cell-derived neurons (Rakovic and others 2013), thus questioning the physiological relevance of this pathway in the mammalian brain in the presence of endogenous levels of PINK1/Parkin.

Mitochondria and Their Close Neighbor, the Endoplasmic Reticulum

Mitochondria do not exist in isolation but interact physically with many other subcellular organelles in a cooperative manner to facilitate rapid and efficient signaling and metabolism. An example of such is their interaction with the endoplasmic reticulum (ER), first reported by

morphological data using electron microscopy (Bernhard and Rouiller 1956). Mitochondria-associated ER membranes, or MAM, display a particular sedimentation profile, composition and enzymatic activity that are different from the bulk ER (Vance 2014). They are enriched in activities related to lipid metabolism, particularly in relation to cholesterol and phospholipid synthesis enzymes and calcium-handling proteins. Nonetheless, ER–mitochondrial contacts are much more than a simple approximation between the two organelles. In effect, they are tethered through different protein interactions, which are reversible and regulated, as evidenced by the interaction between VDAC (voltage-dependent anion-selective channel) and IP3 receptor (Szabadkai and others 2006) or by the enrichment of Mitofusin 2 (MFN2) at the ER-mitochondrion interface. Mitofusin 2 not only participates in mitochondrial fusion but also interacts with MFN1 or MFN2, present on the mitochondrial surface, leading to the aproximation of both organelles (de Brito and Scorrano 2008). The close apposition between mitochondria and ER, together with the selective presence of proteins and enzymes in this region, makes the ER–mitochondrial surface the perfect site for key cellular processes such as lipid transfer and metabolism, calcium transfer/signaling and cell death, to take place (Fig. 5). The accumulation of calcium within mitochondria leads to the activation of oxidative phosphorylation by regulating the activity of three dehydrogenases in the Krebs cycle (Denton 2009); this is crucial for matching ATP production to metabolic demands stemming from neuronal electrical activity. SNpc dopaminergic neurons are autonomously active, with the pacemaking activity of these neurons driven by voltage-dependent L-type calcium channels, leading to sustained elevations in cytosolic calcium concentrations (Chan and others 2007). The large calcium-buffering burden created by pacemaking activity in SNpc dopaminergic neurons ultimately compromises mitochondrial function which may thus underlie the selective vulnerability of SNpc dopaminergic neurons in PD. Indeed, as reviewed elsewhere (Rao and others 2014), calcium overload in the mitochondria favors the opening of the mitochondrial permeability transition pore (PTP), together with cytochrome c release and apoptosis. Szalai and others (1999) showed that IP3 receptor–induced opening of the PTP is dependent on calcium signal transmission from IP3 receptors to the mitochondria. Inside the mitochondria, calcium binds to adenine-nucleotide-translocator, which is thought to be converted into an unselective channel. This leads to the swelling of the inner mitochondrial membrane together with the loss of membrane potential. Another consequence is the breakdown of the outer mitochondrial membrane and, finally, the release of pro-apoptotic factors such as cytochrome c. This is only one example of how this newly identified subdomain in the ER/mitochondria may be the site where classical processes tied to neurodegeneration take place. In fact, proteins such as Drp1 or MFN2, which are linked to neurodegenerative diseases, are enriched in MAM (de Brito and Scorrano 2008). When taken together, one might wonder whether ER-mitochondrial contacts might contribute to the pathogenesis of neurodegenerative diseases.

In the case of PD, it has been reported that α-synuclein is present not only in the cytosol but is also associated with the mitochondria (Stefanis 2012) and that overexpression of α-synuclein leads to an increase of calcium transfer from the ER to the mitochondria, together with an up-regulation of mitochondria–ER contacts in human cells. In contrast, a siRNA-mediated down-regulation of α-synuclein reduces mitochondrial-calcium uptake (Calì and others 2012). More recently, it was shown that α-synuclein is also localized at the MAM (Guardia-Laguarta and others 2014), where it promotes the transfer of calcium from the ER to the mitochondria (Calì and others 2012). Remarkably, PD-linked α-synuclein mutations disrupt the association of α-synuclein with MAM, resulting in a decreased apposition of ER with the mitochondria and a decrease in MAM function (Guardia-Laguarta and others 2014). Furthermore, DJ-1 modulates mitochondrial calcium levels by favoring ER-mitochondria tethering. A reduction of DJ-1 levels results in mitochondria fragmentation and decreased mitochondrial calcium uptake in stimulated cells, suggesting that the impairment of ER-mitochondrial communication, as a consequence of DJ-1 loss-of-function, may play a role in the mitochondrial dysfunction observed in PD (Ottolini and others 2013) (Fig. 5).

Conclusions: From Mitochondrial Dysfunction to Neuroprotection

As mitochondria are central to the regulation of energy production, calcium homeostasis, bioenergetic quality control and cell death regulation, maintaining their functions in an operational manner is thus a priority for the cell. This dependence of cells on the mitochondrial energy production is particularly true for dopaminergic neurons of the SNpc as in humans the brain uses more energy than any other organ (~20% of the body's total demand), and those neurons have as a common characteristic to be long-range projection neurons with an elaborate axonal arborisation and a high rate of mitochondrial oxidative metabolism. It is now well established that mitochondrial dysregulation plays a central pathogenic role in PD. However, to date, there is no clear indication as to whether mitochondrial dysfunction is a cause of PD or instead is correlated with the progression of the disease. If mitochondrial dysfunction is central to the pathogenesis of PD, one could expect to see PD symptoms in patients with mitochondrial disorders. Reeve and others (2013) recently analyzed the effects

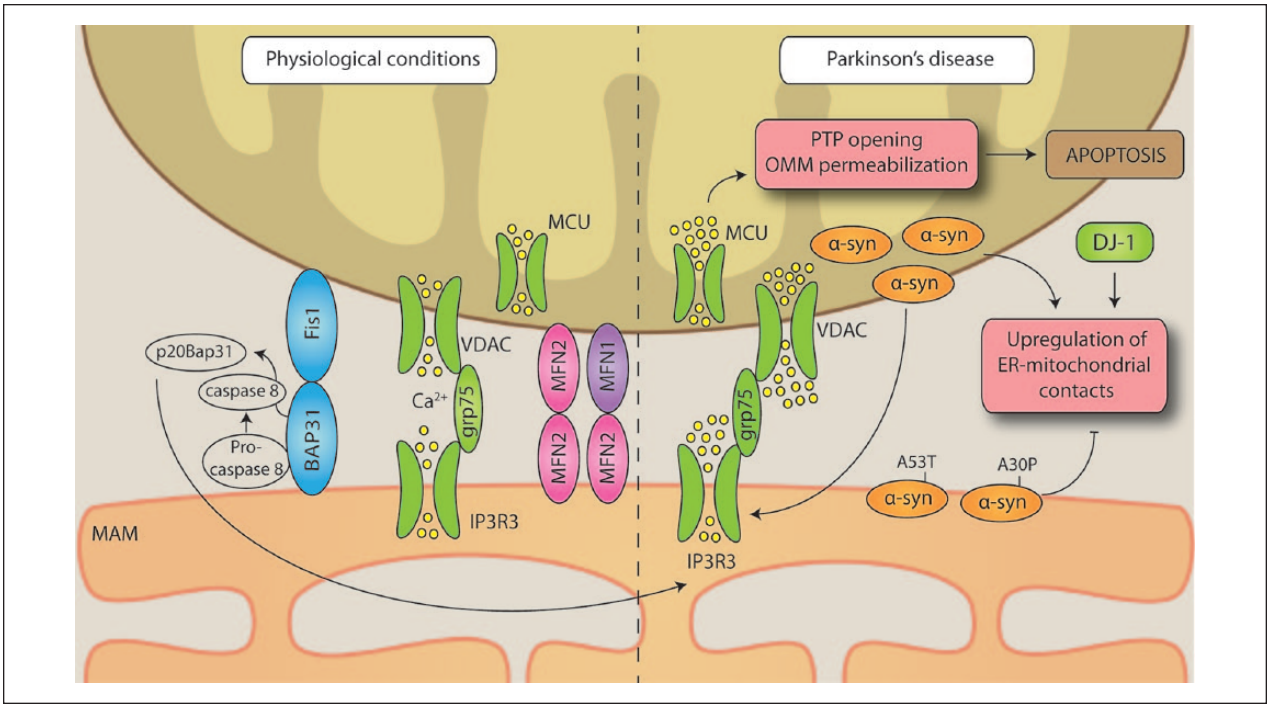

Figure 5. Endoplasmic reticulum (ER)–mitochondrial contacts and Parkinson's disease (PD). The ER–mitochondrial junction is the site where key cellular processes take place. The close apposition between ER and mitochondrial proteins enables the formation of an apoptosis platform in the BAP31-Fis1 interaction. This mediates pro-caspase-8 recruitment and activation, leading to generation of the p20Bap31 apoptotic fragment that activates calcium release from ER stores. Calcium release from inositol-1,4,5-triphosphate receptor 3 (IP3R3) is facilitated because of its interaction with voltage-dependent anionic channels (VDAC) through the molecular chaperone grp75. Another ER-mitochondrial tether is Mitofusin 2 (MFN2), which is also located in mitochondria-associated ER membranes (MAM) and can interact with Mitofusin 1 (MFN1) and MFN2 in the outer mitochondrial membrane. ER-mitochondrial contacts play an important role in the pathogenesis of PD. α-Synuclein (α-syn) overexpression leads to increased calcium transfer due to the up-regulation of ER–mitochondrial contacts, which can result in calcium-dependent apoptosis. Whereas α-syn has been shown to localize to MAM, PD-linked mutations in α-syn disrupt this association, thus decreasing ER–mitochondrial apposition. On the other hand, DJ-1 favors ER-mitochondrial tethering, with reduced DJ-1 levels resulting in decreased mitochondrial calcium uptake. Ca^{2+} = calcium; MCU = mitochondrial calcium uniporter; PTP = permeability transition pore.

of mitochondrial mutations—known to cause cell loss and dysfunction in other brain regions and tissues—on mitochondrial function and dopaminergic neurons in patients. Defects in both complex I and complex IV of the respiratory chain were seen in SNpc neuron mitochondria, but only a subset of patients with mutations in POLG exhibited marked neuronal loss. Why are only a few cases of mitochondrial disease associated with Parkinsonism? Two hypothesis can be advanced: (a) Neurons in which a defect develops early in life are more able to adapt and less likely to degenerate and (b) as dopaminergic cell loss was only observed in some mitochondrial patients with POLG mutations associated with α-synuclein pathology (Reeve and others 2013), it is possible that mitochondrial dysfunction below a given threshold is not sufficient to induce dopaminergic cell death but may be part of a more complex multifactorial pathogenic process.

Different therapeutic strategies were proposed to bypass mitochondrial dysfunctions in experimental PD. These include the "boost" of energy production, the scavenging of free radicals, the inhibition of calcium entry within the cell, the inhibition of mitochondria-dependent PCD, and the inhibition of mitochondrial division. Unfortunately, so far, none of those treatment strategies have provided an ameliorative effect on motor symptoms in patients. Disappointing clinical trial results involving mitochondrial therapies have raised the question of whether the presently used animal models adequately reflect the underlying neurodegenerative processes of idiopathic PD. We strongly believe that even if the existing models are far from being ideal, and the development of new animal models is necessary, the "classical" toxin-based animal models have greatly contributed to the development of current PD treatments such as L-dopa,

dopamine agonists, and monoamine oxidase-B inhibitors, and have increased our understanding of the basal ganglia circuitry, pathogenic mechanisms, and potential therapeutic targets for PD. The actual challenge will be to refine the process of patient selection into clinical trials and to develop early clinical markers for PD. Indeed, it is likely that a number of factors participate in the pathogenesis of the disease and it might be necessary to identify subgroups of patients that may potentially benefit from candidate drugs affecting mitochondrial function (patients with decreased complex I activity, patients with Parkin/PINK1 mutations, etc.). Furthermore, by the time most patients develop the typical clinical symptoms of PD, it is estimated that at least 60% of nigral dopamine neurons have degenerated. For a potential neuroprotective agent to be most effective, patients need to be treated early, and before significant cell loss has occurred. For this it is necessary for early clinical markers for PD to be developed. Despite the challenges lying ahead, we are confident that in the next few years, major breakthroughs relating to mitochondrial therapies will appear.

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References

Andrabi SA, Dawson TM, Dawson VL. 2008. Mitochondrial and nuclear cross talk in cell death: parthanatos. Ann N Y Acad Sci 1147:233–41.

Andres-Mateos E, Perier C, Zhang L, Blanchard-Fillion B, Greco TM, Thomas B, and others. 2007. DJ-1 gene deletion reveals that DJ-1 is an atypical peroxiredoxin-like peroxidase. Proc Natl Acad Sci U S A 104:14807–12.

Anglade P. 1997. Apoptosis and autophagy in nigral neurons of patients with Parkinson's disease. Histol Histopathol 12:25–31.

Bender A, Krishnan KJ, Morris CM, Taylor GA, Reeve AK, Perry RH, and others. 2006. High levels of mitochondrial DNA deletions in substantia nigra neurons in aging and Parkinson disease. Nat Genet 38:515–7.

Bernhard W, Rouiller C. 1956. Close topographical relationship between mitochondria and ergastoplasm of liver cells in a definite phase of cellular activity. J Biophys Biochem Cytol 2:73–8.

Berthet A, Margolis EB, Zhang J, Hsieh I, Zhang J, Hnasko TS, and others. 2014. Loss of mitochondrial fission depletes axonal mitochondria in midbrain dopamine neurons. J Neurosci 34:14304–17.

Betarbet R, Sherer TB, Mackenzie G, Garcia-Osuna M, Panov AV, Greenamyre JT. 2000. Chronic systemic pesticide exposure reproduces features of Parkinson's disease. Nat Neurosci 3:1301–6.

Bonifati V, Rizzu P, van Baren MJ, Schaap O, Breedveld GJ, Krieger E, and others. 2003. Mutations in the DJ-1 gene associated with autosomal recessive early-onset parkinsonism. Science 299:256–9.

Büttner S, Habernig L, Broeskamp F, Ruli D, Vögtle FN, Vlachos M, and others. 2013. Endonuclease G mediates α-synuclein cytotoxicity during Parkinson's disease. EMBO J 32:3041–54.

Calì T, Ottolini D, Negro A, Brini M. 2012. α-Synuclein controls mitochondrial calcium homeostasis by enhancing endoplasmic reticulum–mitochondria interactions. J Biol Chem 287:17914–29.

Chan CS, Guzman JN, Ilijic E, Mercer JN, Rick C, Tkatch T, and others. 2007. "Rejuvenation" protects neurons in mouse models of Parkinson's disease. Nature 447:1081–6.

Chang DTW, Honick AS, Reynolds IJ. 2006. Mitochondrial trafficking to synapses in cultured primary cortical neurons. J Neurosci 26:7035–45.

Charan RA, Johnson BN, Zaganelli S, Nardozzi JD, LaVoie MJ. 2014. Inhibition of apoptotic Bax translocation to the mitochondria is a central function of parkin. Cell Death Dis 5:e1313.

Chu CT, Zhu J, Cao G, Signore A, Wang S, Chen J. 2005. Apoptosis inducing factor mediates caspase-independent 1-methyl-4-phenylpyridinium toxicity in dopaminergic cells. J Neurochem 94:1685–95.

Ciron C, Lengacher S, Dusonchet J, Aebischer P, Schneider BL. 2012. Sustained expression of PGC-1α in the rat nigrostriatal system selectively impairs dopaminergic function. Hum Mol Genet 21:1861–76.

Dai Y, Clark J, Zheng K, Kujoth GC, Prolla TA, Simon DK. 2014. Somatic mitochondrial DNA mutations do not increase neuronal vulnerability to MPTP in young POLG mutator mice. Neurotoxicol Teratol 46C:62–7.

Darios F, Corti O, Lücking CB, Hampe C, Muriel M-P, Abbas N, and others. 2003. Parkin prevents mitochondrial swelling and cytochrome c release in mitochondria-dependent cell death. Hum Mol Genet 12:517–26.

Dauer W, Przedborski S. 2003. Parkinson's disease: mechanisms and models. Neuron 39:889–909.

Davey GP, Clark JB. 1996. Threshold effects and control of oxidative phosphorylation in nonsynaptic rat brain mitochondria. J Neurochem 66:1617–24.

de Brito OM, Scorrano L. 2008. Mitofusin 2 tethers endoplasmic reticulum to mitochondria. Nature 456:605–10.

Dehay B, Martinez-Vicente M, Caldwell GA, Caldwell KA, Yue Z, Cookson MR, and others. 2013. Lysosomal impairment in Parkinson's disease. Mov Disord 28:725–32.

Denton RM. 2009. Regulation of mitochondrial dehydrogenases by calcium ions. Biochim Biophys Acta 1787:1309–16.

Detmer SA, Chan DC. 2007. Functions and dysfunctions of mitochondrial dynamics. Nat Rev Mol Cell Biol 8: 870–9.

Ekstrand MI, Terzioglu M, Galter D, Zhu S, Hofstetter C, Lindqvist E, and others. 2007. Progressive parkinsonism in mice with respiratory-chain-deficient dopamine neurons. Proc Natl Acad Sci U S A 104:1325–30.

Eschbach J, von Einem B, Müller K, Bayer H, Scheffold A, Morrison BE, and others. 2015. Mutual exacerbation of PGC-1α deregulation and α-synuclein oligomerization. Ann Neurol 77:15–32.

Frezza C, Cipolat S, Martins de Brito O, Micaroni M, Beznoussenko GV, Rudka T, and others. 2006. OPA1 controls apoptotic cristae remodeling independently from mitochondrial fusion. Cell 126:177–89.

Guardia-Laguarta C, Area-Gomez E, Rüb C, Liu Y, Magrané J, Becker D, and others. 2014. α-Synuclein is localized to mitochondria-associated ER membranes. J Neurosci 34:249–59.

Hattori N, Tanaka M, Ozawa T, Mizuno Y. 1991. Immunohistochemical studies on complexes I, II, III, and IV of mitochondria in Parkinson's disease. Ann Neurol 30:563–71.

Hudson G, Nalls M, Evans JR, Breen DP, Winder-Rhodes S, Morrison KE, and others. 2013. Two-stage association study and meta-analysis of mitochondrial DNA variants in Parkinson disease. Neurology 80:2042–8.

Iaccarino C, Crosio C, Vitale C, Sanna G, Carrì MT, Barone P. 2007. Apoptotic mechanisms in mutant LRRK2-mediated cell death. Hum Mol Genet. 16:1319–26.

Irrcher I, Aleyasin H, Seifert EL, Hewitt SJ, Chhabra S, Phillips M, and others. 2010. Loss of the Parkinson's disease-linked gene DJ-1 perturbs mitochondrial dynamics. Hum Mol Genet 19:3734–46.

Keeney PM, Xie J, Capaldi RA, Bennett JP. 2006. Parkinson's disease brain mitochondrial complex I has oxidatively damaged subunits and is functionally impaired and misassembled. J Neurosci 26:5256–64.

Krebiehl G, Ruckerbauer S, Burbulla LF, Kieper N, Maurer B, Waak J, and others. 2010. Reduced basal autophagy and impaired mitochondrial dynamics due to loss of Parkinson's disease-associated protein DJ-1. PLoS One 5:e9367.

Kujoth GC, Hiona A, Pugh TD, Someya S, Panzer K, Wohlgemuth SE, and others. 2005. Mitochondrial DNA mutations, oxidative stress, and apoptosis in mammalian aging. Science 309:481–4.

Langston JW, Ballard P, Tetrud JW, Irwin I. 1983. Chronic Parkinsonism in humans due to a product of meperidine-analog synthesis. Science 219:979–80.

Lemasters JJ. 2005. Selective mitochondrial autophagy, or mitophagy, as a targeted defense against oxidative stress, mitochondrial dysfunction, and aging. Rejuvenation Res 8:3–5.

Loeb V, Yakunin E, Saada A, Sharon R. 2010. The transgenic overexpression of α-synuclein and not its related pathology associates with complex I inhibition. J Biol Chem 285:7334–43.

Mandir AS, Przedborski S, Jackson-Lewis V, Wang ZQ, Simbulan-Rosenthal CM, Smulson ME, and others. 1999. Poly(ADP-ribose) polymerase activation mediates 1-methyl-4-phenyl-1, 2,3,6-tetrahydropyridine (MPTP)-induced parkinsonism. Proc Natl Acad Sci U S A 96:5774–9.

Martinat C, Shendelman S, Jonason A, Leete T, Beal MF, Yang L, and others. 2004. Sensitivity to oxidative stress in DJ-1-deficient dopamine neurons: an ES-derived cell model of primary Parkinsonism. PLoS Biol 2:e327.

Mudò G, Mäkelä J, Di Liberto V, Tselykh T V, Olivieri M, Piepponen P, and others. 2012. Transgenic expression and activation of PGC-1α protect dopaminergic neurons in the MPTP mouse model of Parkinson's disease. Cell Mol Life Sci 69:1153–65.

Ottolini D, Calì T, Negro A, Brini M. 2013. The Parkinson disease-related protein DJ-1 counteracts mitochondrial impairment induced by the tumour suppressor protein p53 by enhancing endoplasmic reticulum–mitochondria tethering. Hum Mol Genet 22:2152–68.

Parker WD, Boyson SJ, Parks JK. 1989. Abnormalities of the electron transport chain in idiopathic Parkinson's disease. Ann Neurol 26:719–23.

Perier C, Bender A, García-Arumí E, Melià MJ, Bové J, Laub C, and others. 2013. Accumulation of mitochondrial DNA deletions within dopaminergic neurons triggers neuroprotective mechanisms. Brain 136:2369–78.

Perier C, Bové J, Dehay B, Jackson-Lewis V, Rabinovitch PS, Przedborski S, and others. 2010. Apoptosis-inducing factor deficiency sensitizes dopaminergic neurons to parkinsonian neurotoxins. Ann Neurol 68:184–92.

Perier C, Bové J, Vila M. 2012. Mitochondria and programmed cell death in Parkinson's disease: apoptosis and beyond. Antioxid Redox Signal 16:883–95.

Perier C, Bové J, Wu D-C, Dehay B, Choi D-K, Jackson-Lewis V, and others. 2007. Two molecular pathways initiate mitochondria-dependent dopaminergic neurode-generation in experimental Parkinson's disease. Proc Natl Acad Sci U S A 104:8161–6.

Perier C, Tieu K, Guégan C, Caspersen C, Jackson-Lewis V, Carelli V, and others. 2005. Complex I deficiency primes Bax-dependent neuronal apoptosis through mitochondrial oxidative damage. Proc Natl Acad Sci U S A 102:19126–31.

Perier C, Vila M. 2012. Mitochondrial biology and Parkinson's disease. Cold Spring Harb Perspect Med 2:a009332.

Pickrell AM, Pinto M, Hida A, Moraes CT. 2011. Striatal dysfunctions associated with mitochondrial DNA damage in dopaminergic neurons in a mouse model of Parkinson's disease. J Neurosci 31:17649–58.

Rakovic A, Shurkewitsch K, Seibler P, Grünewald A, Zanon A, Hagenah J, and others. 2013. Phosphatase and tensin homolog (PTEN)-induced putative kinase 1 (PINK1)-dependent ubiquitination of endogenous Parkin attenuates mitophagy: study in human primary fibroblasts and induced pluripotent stem cell-derived neurons. J Biol Chem 288:2223–37.

Ramonet D, Perier C, Recasens A, Dehay B, Bové J, Costa V, and others. 2013. Optic atrophy 1 mediates mitochondria remodeling and dopaminergic neurodegeneration linked to complex I deficiency. Cell Death Differ 20:77–85.

Rao VK, Carlson EA, Yan SS. 2014. Mitochondrial permeability transition pore is a potential drug target for neurodegeneration. Biochim Biophys Acta 1842:1267–72.

Rappold PM, Cui M, Grima JC, Fan RZ, de Mesy-Bentley KL, Chen L, and others. 2014. Drp1 inhibition attenuates neurotoxicity and dopamine release deficits in vivo. Nat Commun 5:5244.

Reeve A, Meagher M, Lax N, Simcox E, Hepplewhite P, Jaros E, and others. 2013. The impact of pathogenic mitochondrial DNA mutations on substantia nigra neurons. J Neurosci 33:10790–801.

Sanders LH, McCoy J, Hu X, Mastroberardino PG, Dickinson BC, Chang CJ, and others. 2014. Mitochondrial DNA damage: molecular marker of vulnerable nigral neurons in Parkinson's disease. Neurobiol Dis 70:214–23.

Santos D, Cardoso SM. 2012. Mitochondrial dynamics and neuronal fate in Parkinson's disease. Mitochondrion 12: 428–37.

Scarffe LA, Stevens DA, Dawson VL, Dawson TM. 2014. Parkin and PINK1: much more than mitophagy. Trends Neurosci 37:315–24.

Schapira AH, Cooper JM, Dexter D, Clark JB, Jenner P, Marsden CD. 1990. Mitochondrial complex I deficiency in Parkinson's disease. J Neurochem 54:823–7.

Stefanis L. 2012. α-Synuclein in Parkinson's disease. Cold Spring Harb Perspect Med 2:a009399.

St-Pierre J, Drori S, Uldry M, Silvaggi JM, Rhee J, Jäger S, and others. 2006. Suppression of reactive oxygen species and neurodegeneration by the PGC-1 transcriptional coactivators. Cell 127:397–408.

Szabadkai G, Bianchi K, Várnai P, De Stefani D, Wieckowski MR, Cavagna D, and others. 2006. Chaperone-mediated coupling of endoplasmic reticulum and mitochondrial Ca^{2+} channels. J Cell Biol 175:901–11.

Szalai G, Krishnamurthy R, Hajnóczky G. 1999. Apoptosis driven by IP(3)-linked mitochondrial calcium signals. EMBO J 18:6349–61.

Twig G, Shirihai OS. 2011. The interplay between mitochondrial dynamics and mitophagy. Antioxid Redox Signal 14:1939–51.

Vance JE. 2014. MAM (mitochondria-associated membranes) in mammalian cells: lipids and beyond. Biochim Biophys Acta 1841:595–609.

Venderova K, Park DS. 2012. Programmed cell death in Parkinson's disease. Cold Spring Harb Perspect Med 2:a009365.

Vives-Bauza C, Przedborski S. 2011. Mitophagy: the latest problem for Parkinson's disease. Trends Mol Med 17: 158–65.

Wang H-L, Chou A-H, Yeh T-H, Li AH, Chen Y-L, Kuo Y-L, and others. 2007. PINK1 mutants associated with recessive Parkinson's disease are defective in inhibiting mitochondrial release of cytochrome c. Neurobiol Dis 28:216–26.

Wang Y, Dawson VL, Dawson TM. 2009. Poly(ADP-ribose) signals to mitochondrial AIF: a key event in parthanatos. Exp Neurol 218:193–202.

Youle RJ, Narendra DP. 2011. Mechanisms of mitophagy. Nat Rev Mol Cell Biol 12:9–14.

Zheng B, Liao Z, Locascio JJ, Lesniak KA., Roderick SS, Watt ML, and others. 2010. PGC-1α, a potential therapeutic target for early intervention in Parkinson's disease. Sci Transl Med 2:52ra73.