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Molecular Mechanisms Mediating Mitochondrial Dynamics and Mitophagy and Their Functional Roles in the Cardiovascular System

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Abstract

Mitochondria are essential organelles that produce the cellular energy source, ATP. Dysfunctional mitochondria are involved in the pathophysiology of heart disease, which is associated with reduced levels of ATP and excessive production of reactive oxygen species. Mitochondria are dynamic organelles that change their morphology through fission and fusion in order to maintain their function. Fusion connects neighboring depolarized mitochondria and mixes their contents to maintain membrane potential. In contrast, fission segregates damaged mitochondria from intact ones, where the damaged part of mitochondria is subjected to mitophagy whereas the intact part to fusion. It is generally believed that mitochondrial fusion is beneficial for the heart, especially under stress conditions, because it consolidates the mitochondria's ability to supply energy. However, both excessive fusion and insufficient fission disrupt the mitochondrial quality control mechanism and potentiate cell death. In this review, we discuss the role of mitochondrial dynamics and mitophagy in the heart and the cardiomyocytes therein, with a focus on their roles in cardiovascular disease.

Keywords

Mitochondria; fusion; fission; mitophagy; Drp1

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DISCLOSURES
None.

Introduction

Mitochondria are essential sources of energy in cells and, thus, are particularly important intracellular organelles in ventricular cardiomyocytes, which require regular frequent contraction. However, mitochondria are also major intracellular sources of reactive oxygen species (ROS), which are produced as byproducts of ATP synthesis through the electron transport chain or through upregulation of ROS producing enzymes, such as Nox4, or downregulation of anti-oxidants. Although ROS produced in mitochondria can act as second messengers to trigger adaptive processes [1-3], mitochondrial damage caused by pathological stress often leads to production of excessive ROS, which develops into a vicious cycle of oxidative stress and mitochondrial damage and spreads rapidly into the intact mitochondria within the same cell through a mechanism known as ROS-induced ROS release [4]. Eventually, the mitochondria release cytochrome c into the cytosol by increasing outer mitochondrial membrane (OMM) permeability and activate apoptosis, a cellular suicide mechanism, in order to avoid a series of catastrophic events. In the presence of severe stress, such as prolonged cardiac ischemia, mitochondrial permeability transition pore (mPTP) opening abrogates the H^+ gradient, which is essential for ATP synthesis, and cells undergo necrosis [5]. Histological analysis shows that the heart contains a large volume of mitochondria, indicating that cardiomyocytes rely heavily upon mitochondrial oxidative metabolism as a source of energy supply [5]. In order to prevent the vicious cycle of mitochondrial damage and ROS production, myocardial cells appears to have intrinsic quality control mechanisms by which they protect themselves from minor injury and maintain their function, such as mitochondrial autophagy [6]. In this review, we discuss the role of mitochondrial dynamics in the cardiovascular system, with a special emphasis on the function of dynamin-related protein 1 (Drp1), a molecule involved in fission and mitophagy, in cardiomyocytes.

Mitochondrial Dynamics: Fission and Fusion

Mitochondria are dynamic organelles that constantly undergo fusion and fission, collectively termed“mitochondrial dynamics”, to adapt to changes in the cellular environment and to maintain their function [6]. Fission produces small spherical mitochondria, whereas fusion produces tubular or elongated-shaped mitochondria [6]. Disruption of mitochondrial fission leads to formation of fused mitochondria, whereas that of fusion leads to formation of small and divided mitochondria, suggesting that the morphological changes in mitochondria are balanced by opposing events. It should be noted that the continuous occurrence of mitochondrial fusion and fission has not been tracked in normal adult ventricular cardiomyocytes and, thus, their roles have been inferred based on pharmacological or genetic manipulation. Although we discuss molecular mechanisms controlling fission and fusion of mitochondria in the following section, almost all works have been conducted using non-cardiac cell types. Thus, caution should be exercised regarding whether the findings from other cell types can be applicable to adult ventricular cardiomyocytes.

Mitochondrial dynamics are regulated by several different guanosine triphosphatases (GTPases), which are well-conserved among yeast, flies, and mammals [7-9]. Mitofusin 1 and 2 (Mfn1 and Mfn2) and optic atrophy 1 (Opa1) are involved in regulating mitochondrial

fusion in the outer and inner mitochondrial membranes, respectively. On the other hand, mitochondrial fission is regulated by mitochondrial fission 1 (Fis1) and mitochondrial fission factor (Mff), localized on the outer mitochondrial membrane, and by recruitment of Drp1 from the cytosol to mitochondrial fission sites, where it interacts with Mff to promote fission [7-9].

Mitochondrial fission is initiated by constriction of mitochondria at points where endoplasmic reticulum (ER) tubules surround mitochondria and mark mitochondrial division sites, which is followed by recruitment of Drp1 to the mitochondria [10]. Although the mechanism by which initial constriction of mitochondria occurs remains unknown, subsequent constriction and scission processes are mediated by Drp1 [11, 12]. Drp1 is an ~80 kDa dynamin GTPase superfamily protein with an N-terminal GTPase domain thought to provide mechanical force, a dynamin-like middle domain, a variable domain, and a C-terminal GTPase effector domain (GED) [13, 14]. Drp1 is abundantly expressed in the skeletal muscle, heart, kidney, and brain of adult humans. Drp1 primarily exists in the cytosol as a dimer/tetramer. During mitochondrial fission, Drp1 translocates from the cytosol to mitochondria, where it oligomerizes around and constricts the mitochondria, thereby leading to severing of the mitochondrial membrane by GTP hydrolysis [15-19]. Drp1 is regulated by a variety of post-translational modifications, including phosphorylation, S-nitrosylation, small ubiquitin-like modifier (SUMO)-ylation, and ubiquitination, in response to diverse cellular stimuli [20-29]. Mitochondrial localization of Drp1 is positively regulated by protein kinase A, calcineurin, PUMA, Bax/Bak, ceramide, and O-linked-β-N-acetylglucosamine (O-GlcNAcylation) modification, and is negatively regulated by miR-499 and Pim1 [28, 30-34]. Drp1 lacks a mitochondrial targeting sequence. Therefore, Drp1-mediated mitochondrial fission requires a receptor to promote Drp1 recruitment to the outer mitochondrial membrane [35, 36]. Fis1 is a 17-kDa protein that anchors to the outer mitochondrial membrane with its N-terminal multiple tetratricopeptide repeat motif exposed to the cytoplasm and was thought to serve as a receptor for Drp1 [37]. Indeed, overexpression of Fis1 in cells promotes mitochondrial fission. However, Fis1 knockdown affects neither recruitment of Drp1 to mitochondria nor fission in HeLa cells and HCT 116 cells [38]. Mff is a C tail-anchored protein [39]. In cells with Mff knockdown, mitochondrial localization of Drp1 is decreased and Drp1 is dispersed in the cytoplasm. In contrast, Mff overexpression induces mitochondrial fission with increased Drp1 recruitment to mitochondria, suggesting that Mff acts as a Drp1 receptor to promote mitochondrial fission [38].

Mitochondrial fusion is regulated by GTPase dynamin-family proteins, including Mfn1, Mfn2, and Opa1 [40, 41]. Mfn1 and Mfn2 localize to the outer mitochondrial membrane and share approximately 77% sequence similarity. Mfn1 and Mfn2 have both redundant and distinct functions to promote mitochondrial fusion by forming either homotypic or heterotypic complexes [42]. Downregulation of Mfn1 or Mfn2 shows fragmentation of mitochondria in MEF cells, suggesting that Mfn1 and Mfn2 have non-redundant functions to promote mitochondrial fusion [42]. It should be noted that although cardiac-specific Mfn1 KO mice exhibited fragmented mitochondria [43], cardiac-specific Mfn2 KO mice exhibited enlarged mitochondria in the heart [44, 45], suggesting that Mfn1 and Mfn2 have distinct roles in regulating mitochondrial fusion in the mouse heart. Mfn2 also regulates shape and

function of the endoplasmic/sarcoplasmic reticulum [46, 47] . Mfn2 plays an important role in mediating autophagosome-lysosome fusion in cardiomyocytes [45]. Furthermore, Mfn2 is phosphorylated by PTEN-induced kinase 1 (PINK1) and serves as a receptor for Parkin during mitophagy [48].

Opa1 is a dynamin-related protein that localizes to and tethers the inner mitochondrial membrane to maintain the integrity of the cristae. Overexpression of Opa1 promotes the formation of a branched network of elongated mitochondria, whereas downregulation of Opa1 induces fragmentation of mitochondria and disorganization of the cristae structures [49, 50]. Opa1 cannot tubulate and fuse mitochondria lacking the outer membrane protein Mfn1, but that is not the case in those lacking Mfn2 [41]. Decreases in the electrochemical potential across the inner mitochondrial membrane or proapoptotic stimuli induce paraplegin-dependent proteolytic cleavage of Opa1, thereby stimulating mitochondrial fragmentation [51].

Physiological Role of Mitochondrial Dynamics

Mitochondrial dynamics are crucial for compensating for mitochondrial damage and for eliminating mitochondria with unrecoverable damage through fusion and fission, respectively. The physiological role of fission is believed to be segregation of unrecoverable damaged mitochondria in order to maintain overall mitochondrial quality and to preserve the health of the mitochondrial network [6, 52]. Mitochondrial fission divides the mitochondrion into functionally uneven daughter mitochondria. In order to achieve asymmetric separation of mitochondria, there may be a sorting event preceding fission, but molecular mechanisms mediating the sorting are currently unknown. The daughter mitochondrion with a normal membrane potential can undergo fusion with other mitochondria. However, the daughter mitochondrion with decreased membrane potential is unable to fuse with other mitochondria, resulting in elimination by mitophagy (Figure 1) [52, 53]. Mitochondrial fission is also necessary to redistribute mitochondrial DNA and transport mitochondria to daughter cells during mitosis [54]. Drp1-mediated fragmentation of mitochondria protects HeLa cells from ceramide-induced, Ca^{2+}-mediated apoptosis, suggesting that fission may segregate intact mitochondria from dysfunctional ones [55].

Fusion is a process by which neighboring depolarized mitochondria and intact ones join together and mix their metabolites and mitochondrial DNA, which allows maintenance of the membrane potential, complementation of protein components, and mtDNA repair [40, 56, 57]. Mitochondrial fusion also maximizes the capacity of oxidative phosphorylation during energy deprivation [58-61]. Therefore, fusion allows mitochondria to compensate for one another's defects. Conversely, impaired mitochondrial fusion decreases overall membrane potential, oxygen consumption and mtDNA replication. It should be noted that fusion may also deteriorate the function of intact mitochondria when depolarized and dysfunctional mitochondria are accumulated in cells due to, for example, impairment of the elimination process [62]. Unopposed fusion induced by inhibition of fission induces mitochondrial dysfunction in HeLa cells [63] and potentiates mPTP opening in MEF cells [64], suggesting that excessive fusion can be detrimental.

Mitochondrial fusion also allows mitochondria to escape elimination by mitophagy [58, 65]. Dominant negative Drp1 or Mfn1 prevents mitochondrial fission and autophagy in cardiomyocytes [66]. However, the exact molecular mechanism preventing autophagy remains unclear. Whether excessive activation of mitophagy could become harmful for the heart and whether the heart purposefully activates mitochondrial fusion to prevent mitophagy remain to be elucidated

Elimination of damaged mitochondria by autophagy

Autophagy is the principal mechanism of cell homeostasis. Autophagy, derived from the Greek words for self (auto) and eating (phagy), is an evolutionarily conserved mechanism of degradation wherein damaged or long-lived proteins and organelles are sequestered into autophagosomes and degraded by lysosomes in order to maintain cell homeostasis. Mitochondria can be degraded by autophagy in either a non-selective manner or through a process that selectively targets damaged mitochondria, termed“mitophagy” [52, 67].

Recent studies have demonstrated that mitophagy is regulated by several specific proteins, including PINK1 and Parkin, which are associated with familial Parkinson's disease [67-70]. PINK1 is a mitochondrially targeted serine/threonine kinase that is imported into healthy mitochondria and degraded by the presenilin-associated rhomboid-like (PARL) protease. Upon mitochondrial depolarization, PINK1 is stabilized and accumulates on the outer membrane of depolarized mitochondria, leading to the recruitment of cytosolic Parkin to mitochondria through phosphorylation of Mfn2 [48]. Parkin is a cytosolic E3-ubiquitin ligase that translocates to depolarized mitochondria and ubiquitinates them. Mitochondria ubiquitinated by Parkin interact with the ubiquitin-binding deacetylase HDAC6 and p62/SQSTM1, which connect with LC3 and promote the assembly of the autophagic machinery to eliminate damaged mitochondria by mitophagy (Figure 2A). As described previously, fission can divide a single mitochondrion into unequal daughter mitochondria with different mitochondrial membrane potentials. Since the PINK1/Parkin pathway depends on low mitochondrial membrane potential, fission may contribute to the targeting of damaged mitochondria to be eliminated by PINK1/Parkin-mediated mitophagy. Interestingly, damaged long mitochondrial tubules labeled by Parkin can be packaged into smaller sized LC3 structures bit-by-bit and undergo mitophagy in HeLa cells [71]. Whether fission is prerequisite for mitophagy and, if so, what the exact role of fission could be remain to be elucidated. Mice deficient in Parkin exhibit impaired clearance of damaged mitochondria by mitophagy and increased accumulation of dysfunctional mitochondria, which results in cardiac dysfunction and reduced survival [72]. Similarly, Pink1 knockout mice exhibited pathological hypertrophy with impaired mitochondrial function at baseline and increased myocardial injury in response to ischemia/reperfusion [73, 74].

Other specific proteins that regulate mitophagy include Bcl-2/adenovirus E1B 19-kDa–interacting protein-3 (Bnip3) and Nip3-like protein X (NIX) [75]. Bnip3 and NIX are pro-apoptotic BH3-only proteins that facilitate opening of the mPTP and activate Bax/Bak to permeabilize the mitochondrial membrane [76, 77]. Besides their roles as pro-apoptotic cell death proteins, they are involved in mitophagy under specific conditions. NIX is required for the selective elimination of mitochondria in erythrocytes in peripheral blood [77]. Although

Bnip3 induces apoptosis in the heart in response to ischemia/reperfusion, it also upregulates autophagy and removal of damaged mitochondria, which appears to be protective [78]. The precise mechanisms by which NIX and Bnip3 induce mitophagy are still unclear. However, Bnip3 and NIX interact directly with LC3 and GABARAP on the phagophore, serving as receptors for autophagosomes that tether them to mitochondria (Figure 2B) [77, 79]. Bnip3 and NIX have been implicated in the pathogenesis of heart disease [66, 80]. Bnip3/NIX-deficient mice exhibit increased accumulation of dysfunctional mitochondria in the heart with age [80]. In addition, inhibition of fission through overexpression of a dominant-negative form of Drp1 leads to disruption of mitophagy induced by Bnip3 and facilitates accumulation of dysfunctional mitochondria in the heart, suggesting that fission is a prerequisite for Bnip3-induced mitophagy [66]. FUNDC1, a mitochondrial outer membrane protein, and cardiolipin, a phospholipid of the inner mitochondrial membrane, also act as LC3 receptors and are involved in mitochondrial autophagy in non-cardiac cell types [81, 82]. Whether mitochondrial shape affects their interaction with LC3 is not well understood.

Mitochondrial Dynamics, Mitophagy and Cardiovascular Disease

Disruption of the mitochondrial quality control mechanisms involving mitochondrial dynamics and mitophagy has recently been linked to various cardiac diseases, including cardiac hypertrophy, heart failure, dilated cardiomyopathy (DCM), and ischemic heart disease. The end stage of DCM, which is characterized by systolic dysfunction and dilated ventricles, is associated with abnormally enhanced fragmentation of mitochondria [83]. Small and fragmented mitochondria are also observed in post-myocardial infarction rat hearts, which are associated with decreased protein levels of Opa1 [84]. Cardiac-specific combined downregulation of Mfn1 and Mfn2 (c-Mfn1/2-KO) induces rapid development of cardiac dysfunction [85, 86], indicating that inhibition of fusion and/or induction of unopposed fission of mitochondria may induce cardiac dysfunction. Similarly, downregulation of Mfn2 stimulates mitochondrial depolarization and release of cytochrome c, resulting in increases in cell death in cardiomyocytes [33]. A potential mechanism through which mitochondrial fission is stimulated during heart failure is Ca^{2+} overload. Increased Ca^{2+} leads to rapid and transient mitochondrial fragmentation and increases in ROS [87]. Python mice carrying a missense mutation in the M domain of Drp1 (C452F) have reduced mitochondrial function, ATP depletion, and consequent energy deficiency, and develop dilated cardiomyopathy [88]. Fibroblasts obtained from Python mice show abnormal mitochondria and peroxisomes. Although the C452F mutation appears to affect the function of Drp1 and the balance between mitochondrial fission and fusion, exactly how it mediates the associated cardiac phenotype remains to be elucidated.

It should be noted that preventing mitochondrial fission through downregulation of Drp1 leads to a loss of mitochondrial DNA, a decrease in mitochondrial respiration and an increase in the abundance of ROS in HeLa cells [63]. Mitochondrial dysfunction resulting from the lack of fission led to a drop in the cellular ATP level, an inhibition of cell proliferation, and an increase in autophagy [63]. These results suggest that mitochondrial remodeling through both fission and fusion may be required for both preservation of mitochondrial function and maintenance of cellular homeostasis in the heart.

Myocardial ischemia not only decreases cellular ATP content but also induces oxidative stress [89], which, in turn, negatively affects function and induces damage to mitochondria. HL-1 cardiomyocytes exhibit mitochondrial fragmentation and mitochondrial dysfunction after 30 minutes of hypoxia followed by normoxia. In this condition, overexpression of either Mfn1 or Mfn2 or expression of a dominant-negative form of Drp1 increased the cell population with elongated mitochondria, decreased mPTP opening, and reduced cell death [90]. Inhibition of Drp1 in HL-1 cells via mitochondrial division inhibitor-1 (mdivi-1), a pharmacological inhibitor of Drp1, reduced mitochondrial dysfunction and damage following hypoxia/normoxia [90]. Furthermore, transient treatment with mdivi-1 reduced myocardial infarct size in mice subjected to 30 minutes of coronary artery occlusion followed by reperfusion [90]. These studies suggest that mitochondrial fission is detrimental and that opposed fusion has protective effects on the heart and the cardiomyocytes therein when afflicted with ischemia/reperfusion (I/R). However, considering that both mitochondrial fusion and fission are essential for mitochondrial remodeling, which plays a crucial role in maintaining mitochondrial quality, it is plausible that prolonged inhibition of fission could be detrimental in cardiac I/R injury. Further investigation is required in order to clarify whether prolonged inhibition of mitochondrial fission is protective for the heart. A recent study showed that stimulation of cultured adult cardiomyocytes with the α-agonist phenylephrine upregulates Drp1 and Fis1, whereas it downregulates Mfn2 [91]. Whether these changes cause mitochondrial remodeling and how they affect the function of mitochondria during cardiac hypertrophy remain to be elucidated.

Endothelial dysfunction contributes to the development of atherosclerosis in patients with diabetes mellitus, leading to increased susceptibility to ischemic heart disease and heart failure. Recent studies have shown that altered mitochondrial dynamics, such as increased mitochondrial fragmentation and Fis1 protein expression, were observed in venous endothelial cells isolated from patients with diabetes mellitus. Exposing cultured human aortic endothelial cells to high levels of glucose yielded increased expression of Fis1 and Drp1 and a loss of mitochondrial networks, which were accompanied by increased mitochondrial ROS production and an impairment of agonist-stimulated activation of endothelial nitric oxide synthase (eNOS) and cGMP production. Knockdown of Fis1 or Drp1 blunted high-glucose-induced alterations in mitochondrial dynamics, ROS production and eNOS activation. These results suggest that increased mitochondrial fission contributes to the development of endothelial dysfunction in diabetic conditions [92]. Mitochondrial fission and Drp1 are also involved in the pathogenesis of pulmonary arterial hypertension (PAH) [93]. PAH is an intractable disease that induces right ventricular failure and is characterized by pulmonary vascular obstruction caused by hyperproliferation of pulmonary artery smooth muscle cells (PASMCs). Activation of hypoxia-inducible factor-1α (HIF-1α) was observed in PASMCs with PAH, which led to mitochondrial fission through cyclin B1/CDK1-dependent phosphorylation of Drp1 at serine 616. HIF-1α inhibition reduced Drp1 activation, prevented mitochondrial fission, and reduced PASMC proliferation. Both mdivi-1 and downregulation of Drp1 prevented mitotic fission and arrested PASMCs at the G2/M interphase. Mdivi-1 also improved exercise capacity, right ventricular function and hemodynamics in experimental PAH, suggesting that Drp1 activation followed by increased mitochondrial fission is responsible for PAH [93]. These results suggest that the status of

mitochondrial remodeling correlates with the function of vascular endothelial cells and smooth muscle cells.

Perspectives

Although acute suppression of mitochondrial fission by mdivi-1 attenuates myocardial injury after I/R [90], unopposed fusion induced by mdivi-1 also induces cell death in other instances [63, 64]. Considering that mitochondrial health is maintained by elimination of damaged and/or malfunctioning mitochondria, the ability of the cell to serially execute fusion and fission of mitochondrial followed by mitophagy appears essential for its survival. In fact, chronic downregulation of Drp1 in neuronal-specific Drp1 knockout mice induces abnormality in embryonic development and synaptic formation, most likely due to defects in mitochondrial quality control mechanisms [94, 95]. Likewise, a deficiency of mitochondrial fission due to Drp1 inactivation is involved in human neurodegenerative pathologies such as Alzheimer's disease [27, 96]. These results suggest that maintaining certain levels of mitochondrial fission may be essential for the survival of cardiomyocytes under some conditions. How do we reconcile the potentially dichotomous effects of mitochondrial fission? Cells may require basal levels of fission but stress-induced activation of fission may be detrimental. Alternatively, transient increases in mitochondrial fusion achieved by pharmacological inhibition of Drp1 may increase mitochondrial function, whereas chronic inhibition of Drp1 by genetic manipulations may lead to the impairment of mitochondrial quality control mechanisms, resulting in global suppression of mitochondrial function. It is also possible that the effect of mitochondrial fission upon mitochondrial function may differ depending upon the nature of the stress. Further investigations are needed to elucidate the functional consequences of mitochondrial fission in response to many forms of cardiac stress. Many of the studies evaluating the role of Drp1 and fission in mediating survival and death of cardiomyocytes have been conducted using a pharmacologic inhibitor of Drp1, mdivi-1, P110, and Dynasore (Table 1). Although mdivi-1 is believed to be a selective inhibitor of Drp1 [97], it performs Drp1-independent actions as well, such as affecting delayed rectifier K+ channels [98]. In addition, Drp1 has diverse functions that are possibly mediated independently of its effects upon fission. For example, mdivi-1 blocks Bax/Bak-dependent release of both Smac/Diablo and cytochrome c in HeLa cells [97]. Thus, caution should be exercised when interpreting the results obtained using inhibitors of Drp1. Ideally, parallel experiments should be conducted, using molecular tools including knock-down of Drp1 and/or, more broadly, interventions to inhibit mitochondrial fission with alternative methods, such as suppression of Fis1 or Mff and stimulation of Mfn1/2 and Opa1.

Mitophagy is an essential mechanism for mitochondrial quality control and it is believed that mitochondrial fission is prerequisite for activation of mitophagy [6]. At present, however, how translocation of Drp1 from the cytosol to the fission foci and subsequent execution of fission coordinate with the activation of mitophagy is not fully understood. Although PINK1-induced phosphorylation of Mfn2 recruits Parkin to damaged mitochondria [48] and consequent ubiquitin labeling of mitochondria with the E3 ligase activity of Parkin is essential for activation of mitophagy [6], whether Drp1 is actively involved in either the labeling process of damaged mitochondria or the sequestration process by autophagic machinery remains to be demonstrated in cardiomyocytes. Similarly, although Drp1 and

mitochondrial fission are coupled to apoptosis in other cell types [97] and the possible involvement of Mfn2 in regulation of mPTP opening has been proposed in cardiomyocytes [43], the molecular mechanisms through which Drp1 affects the activity of the apoptotic machinery are not fully understood in cardiomyocytes.

In summary, mitochondrial fission and Drp1 appear to affect survival and death of cardiomyocytes directly through suppression of mitochondrial fusion and indirectly through their effects upon mitophagy and apoptosis. Further investigations are required to elucidate the molecular mechanism by which mitochondrial fission and Drp1 contribute to the pathogenesis of cardiovascular disease, as well as how interventions targeting Drp1 affect it.

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References

1. Song M, Chen Y, Gong G, Murphy E, Rabinovitch PS, Dorn GW 2nd. Super-suppression of mitochondrial reactive oxygen species signaling impairs compensatory autophagy in primary mitophagic cardiomyopathy. Circ Res. 2014; 115:348–53. [PubMed: 24874428]
2. Yuan H, Perry CN, Huang C, Iwai-Kanai E, Carreira RS, Glembotski CC, et al. LPS-induced autophagy is mediated by oxidative signaling in cardiomyocytes and is associated with cytoprotection. Am J Physiol Heart Circ Physiol. 2009; 296:H470–9. [PubMed: 19098111]
3. Yun J, Finkel T. Mitohormesis. Cell Metab. 2014; 19:757–66. [PubMed: 24561260]
4. Zorov DB, Juhaszova M, Sollott SJ. Mitochondrial reactive oxygen species (ROS) and ROS-induced ROS release. Physiol Rev. 2014; 94:909–50. [PubMed: 24987008]
5. Gottlieb RA, Gustafsson AB. Mitochondrial turnover in the heart. Biochim Biophys Acta. 2011; 1813:1295–301. [PubMed: 21147177]
6. Youle RJ, van der Bliek AM. Mitochondrial fission, fusion, and stress. Science. 2012; 337:1062–5. [PubMed: 22936770]
7. Liesa M, Palacin M, Zorzano A. Mitochondrial dynamics in mammalian health and disease. Physiol Rev. 2009; 89:799–845. [PubMed: 19584314]
8. Otera H, Mihara K. Molecular mechanisms and physiologic functions of mitochondrial dynamics. J Biochem. 2011; 149:241–51. [PubMed: 21233142]
9. Suen DF, Norris KL, Youle RJ. Mitochondrial dynamics and apoptosis. Genes Dev. 2008; 22:1577–90. [PubMed: 18559474]
10. Friedman JR, Lackner LL, West M, DiBenedetto JR, Nunnari J, Voeltz GK. ER tubules mark sites of mitochondrial division. Science. 2011; 334:358–62. [PubMed: 21885730]
11. Frank S, Gaume B, Bergmann-Leitner ES, Leitner WW, Robert EG, Catez F, et al. The role of dynamin-related protein 1, a mediator of mitochondrial fission, in apoptosis. Dev Cell. 2001; 1:515–25. [PubMed: 11703942]
12. Labrousse AM, Zappaterra MD, Rube DA, van der Bliek AM. C. elegans dynamin-related protein DRP-1 controls severing of the mitochondrial outer membrane. Mol Cell. 1999; 4:815–26. [PubMed: 10619028]
13. Elgass K, Pakay J, Ryan MT, Palmer CS. Recent advances into the understanding of mitochondrial fission. Biochim Biophys Acta. 2013; 1833:150–61. [PubMed: 22580041]
14. Otera H, Ishihara N, Mihara K. New insights into the function and regulation of mitochondrial fission. Biochim Biophys Acta. 2013; 1833:1256–68. [PubMed: 23434681]

15. Ingerman E, Perkins EM, Marino M, Mears JA, McCaffery JM, Hinshaw JE, et al. Dnm1 forms spirals that are structurally tailored to fit mitochondria. J Cell Biol. 2005; 170:1021–7. [PubMed: 16186251]
16. Lackner LL, Horner JS, Nunnari J. Mechanistic analysis of a dynamin effector. Science. 2009; 325:874–7. [PubMed: 19679814]
17. Legesse-Miller A, Massol RH, Kirchhausen T. Constriction and Dnm1p recruitment are distinct processes in mitochondrial fission. Mol Biol Cell. 2003; 14:1953–63. [PubMed: 12802067]
18. Mears JA, Lackner LL, Fang S, Ingerman E, Nunnari J, Hinshaw JE. Conformational changes in Dnm1 support a contractile mechanism for mitochondrial fission. Nature structural & molecular biology. 2011; 18:20–6.
19. Smirnova E, Griparic L, Shurland DL, van der Bliek AM. Dynamin-related protein Drp1 is required for mitochondrial division in mammalian cells. Mol Biol Cell. 2001; 12:2245–56. [PubMed: 11514614]
20. Braschi E, Zunino R, McBride HM. MAPL is a new mitochondrial SUMO E3 ligase that regulates mitochondrial fission. EMBO Rep. 2009; 10:748–54. [PubMed: 19407830]
21. Cho DH, Nakamura T, Fang J, Cieplak P, Godzik A, Gu Z, et al. S-nitrosylation of Drp1 mediates beta-amyloid-related mitochondrial fission and neuronal injury. Science. 2009; 324:102–5. [PubMed: 19342591]
22. Figueroa-Romero C, Iniguez-Lluhi JA, Stadler J, Chang CR, Arnoult D, Keller PJ, et al. SUMOylation of the mitochondrial fission protein Drp1 occurs at multiple nonconsensus sites within the B domain and is linked to its activity cycle. FASEB J. 2009; 23:3917–27. [PubMed: 19638400]
23. Horn SR, Thomenius MJ, Johnson ES, Freel CD, Wu JQ, Coloff JL, et al. Regulation of mitochondrial morphology by APC/CCdh1-mediated control of Drp1 stability. Mol Biol Cell. 2011; 22:1207–16. [PubMed: 21325626]
24. Karbowski M, Neutzner A, Youle RJ. The mitochondrial E3 ubiquitin ligase MARCH5 is required for Drp1 dependent mitochondrial division. J Cell Biol. 2007; 178:71–84. [PubMed: 17606867]
25. Kim H, Scimia MC, Wilkinson D, Trelles RD, Wood MR, Bowtell D, et al. Fine-tuning of Drp1/Fis1 availability by AKAP121/Siah2 regulates mitochondrial adaptation to hypoxia. Mol Cell. 2011; 44:532–44. [PubMed: 22099302]
26. Nakamura T, Cieplak P, Cho DH, Godzik A, Lipton SA. S-nitrosylation of Drp1 links excessive mitochondrial fission to neuronal injury in neurodegeneration. Mitochondrion. 2010; 10:573–8. [PubMed: 20447471]
27. Wang H, Song P, Du L, Tian W, Yue W, Liu M, et al. Parkin ubiquitinates Drp1 for proteasome-dependent degradation: implication of dysregulated mitochondrial dynamics in Parkinson disease. J Biol Chem. 2011; 286:11649–58. [PubMed: 21292769]
28. Wasiak S, Zunino R, McBride HM. Bax/Bak promote sumoylation of DRP1 and its stable association with mitochondria during apoptotic cell death. J Cell Biol. 2007; 177:439–50. [PubMed: 17470634]
29. Yonashiro R, Ishido S, Kyo S, Fukuda T, Goto E, Matsuki Y, et al. A novel mitochondrial ubiquitin ligase plays a critical role in mitochondrial dynamics. EMBO J. 2006; 25:3618–26. [PubMed: 16874301]
30. Cribbs JT, Strack S. Reversible phosphorylation of Drp1 by cyclic AMP-dependent protein kinase and calcineurin regulates mitochondrial fission and cell death. EMBO Rep. 2007; 8:939–44. [PubMed: 17721437]
31. Din S, Mason M, Volkers M, Johnson B, Cottage CT, Wang Z, et al. Pim-1 preserves mitochondrial morphology by inhibiting dynamin-related protein 1 translocation. Proc Natl Acad Sci U S A. 110:5969–74. [PubMed: 23530233]
32. Gawlowski T, Suarez J, Scott B, Torres-Gonzalez M, Wang H, Schwappacher R, et al. Modulation of dynamin-related protein 1 (DRP1) function by increased O-linked-beta-N-acetylglucosamine modification (O-GlcNAc) in cardiac myocytes. J Biol Chem. 2012; 287:30024–34. [PubMed: 22745122]

33. Parra V, Eisner V, Chiong M, Criollo A, Moraga F, Garcia A, et al. Changes in mitochondrial dynamics during ceramide-induced cardiomyocyte early apoptosis. Cardiovasc Res. 2008; 77:387–97. [PubMed: 18006463]
34. Wang JX, Jiao JQ, Li Q, Long B, Wang K, Liu JP, et al. miR-499 regulates mitochondrial dynamics by targeting calcineurin and dynamin-related protein-1. Nat Med. 2011; 17:71–8. [PubMed: 21186368]
35. James DI, Parone PA, Mattenberger Y, Martinou JC. hFis1, a novel component of the mammalian mitochondrial fission machinery. J Biol Chem. 2003; 278:36373–9. [PubMed: 12783892]
36. Yoon Y, Krueger EW, Oswald BJ, McNiven MA. The mitochondrial protein hFis1 regulates mitochondrial fission in mammalian cells through an interaction with the dynamin-like protein DLP1. Mol Cell Biol. 2003; 23:5409–20. [PubMed: 12861026]
37. Suzuki M, Jeong SY, Karbowski M, Youle RJ, Tjandra N. The solution structure of human mitochondria fission protein Fis1 reveals a novel TPR-like helix bundle. J Mol Biol. 2003; 334:445–58. [PubMed: 14623186]
38. Otera H, Wang C, Cleland MM, Setoguchi K, Yokota S, Youle RJ, et al. Mff is an essential factor for mitochondrial recruitment of Drp1 during mitochondrial fission in mammalian cells. J Cell Biol. 2010; 191:1141–58. [PubMed: 21149567]
39. Gandre-Babbe S, van der Bliek AM. The novel tail-anchored membrane protein Mff controls mitochondrial and peroxisomal fission in mammalian cells. Mol Biol Cell. 2008; 19:2402–12. [PubMed: 18353969]
40. Chen H, Chan DC. Emerging functions of mammalian mitochondrial fusion and fission. Hum Mol Genet. 2005; 14:R283–9. Spec No. 2. [PubMed: 16244327]
41. Cipolat S, Martins de Brito O, Dal Zilio B, Scorrano L. OPA1 requires mitofusin 1 to promote mitochondrial fusion. Proc Natl Acad Sci U S A. 2004; 101:15927–32. [PubMed: 15509649]
42. Chen H, Detmer SA, Ewald AJ, Griffin EE, Fraser SE, Chan DC. Mitofusins Mfn1 and Mfn2 coordinately regulate mitochondrial fusion and are essential for embryonic development. J Cell Biol. 2003; 160:189–200. [PubMed: 12527753]
43. Papanicolaou KN, Phillippo MM, Walsh K. Mitofusins and the mitochondrial permeability transition: the potential downside of mitochondrial fusion. Am J Physiol Heart Circ Physiol. 2012; 303:H243–55. [PubMed: 22636681]
44. Papanicolaou KN, Khairallah RJ, Ngoh GA, Chikando A, Luptak I, O'Shea KM, et al. Mitofusin-2 maintains mitochondrial structure and contributes to stress-induced permeability transition in cardiac myocytes. Mol Cell Biol. 2011; 31:1309–28. [PubMed: 21245373]
45. Zhao T, Huang X, Han L, Wang X, Cheng H, Zhao Y, et al. Central role of mitofusin 2 in autophagosome-lysosome fusion in cardiomyocytes. J Biol Chem. 2012; 287:23615–25. [PubMed: 22619176]
46. Legros F, Lombes A, Frachon P, Rojo M. Mitochondrial fusion in human cells is efficient, requires the inner membrane potential, and is mediated by mitofusins. Mol Biol Cell. 2002; 13:4343–54. [PubMed: 12475957]
47. Chen Y, Csordas G, Jowdy C, Schneider TG, Csordas N, Wang W, et al. Mitofusin 2-containing mitochondrial-reticular microdomains direct rapid cardiomyocyte bioenergetic responses via interorganelle Ca(2+) crosstalk. Circ Res. 2012; 111:863–75. [PubMed: 22777004]
48. Chen Y, Dorn GW 2nd. PINK1-phosphorylated mitofusin 2 is a Parkin receptor for culling damaged mitochondria. Science. 2013; 340:471–5. [PubMed: 23620051]
49. Griparic L, van der Wel NN, Orozco IJ, Peters PJ, van der Bliek AM. Loss of the intermembrane space protein Mgm1/OPA1 induces swelling and localized constrictions along the lengths of mitochondria. J Biol Chem. 2004; 279:18792–8. [PubMed: 14970223]
50. Olichon A, Baricault L, Gas N, Guillou E, Valette A, Belenguer P, et al. Loss of OPA1 perturbates the mitochondrial inner membrane structure and integrity, leading to cytochrome c release and apoptosis. J Biol Chem. 2003; 278:7743–6. [PubMed: 12509422]
51. Ishihara N, Fujita Y, Oka T, Mihara K. Regulation of mitochondrial morphology through proteolytic cleavage of OPA1. EMBO J. 2006; 25:2966–77. [PubMed: 16778770]

52. Twig G, Elorza A, Molina AJ, Mohamed H, Wikstrom JD, Walzer G, et al. Fission and selective fusion govern mitochondrial segregation and elimination by autophagy. Embo J. 2008; 27:433–46. [PubMed: 18200046]

53. Mouli PK, Twig G, Shirihai OS. Frequency and selectivity of mitochondrial fusion are key to its quality maintenance function. Biophysical journal. 2009; 96:3509–18. [PubMed: 19413957]

54. Hales KG. The machinery of mitochondrial fusion, division, and distribution, and emerging connections to apoptosis. Mitochondrion. 2004; 4:285–308. [PubMed: 16120392]

55. Szabadkai G, Simoni AM, Chami M, Wieckowski MR, Youle RJ, Rizzuto R. Drp-1-dependent division of the mitochondrial network blocks intraorganellar Ca2+ waves and protects against Ca2+-mediated apoptosis. Mol Cell. 2004; 16:59–68. [PubMed: 15469822]

56. Nakada K, Inoue K, Hayashi J. Interaction theory of mammalian mitochondria. Biochem Biophys Res Commun. 2001; 288:743–6. [PubMed: 11688969]

57. Twig G, Hyde B, Shirihai OS. Mitochondrial fusion, fission and autophagy as a quality control axis: the bioenergetic view. Biochim Biophys Acta. 2008; 1777:1092–7. [PubMed: 18519024]

58. Gomes LC, Di Benedetto G, Scorrano L. During autophagy mitochondria elongate, are spared from degradation and sustain cell viability. Nat Cell Biol. 2011; 13:589–98. [PubMed: 21478857]

59. Rambold AS, Kostelecky B, Elia N, Lippincott-Schwartz J. Tubular network formation protects mitochondria from autophagosomal degradation during nutrient starvation. Proc Natl Acad Sci U S A. 2011; 108:10190–5. [PubMed: 21646527]

60. Rossignol R, Gilkerson R, Aggeler R, Yamagata K, Remington SJ, Capaldi RA. Energy substrate modulates mitochondrial structure and oxidative capacity in cancer cells. Cancer Res. 2004; 64:985–93. [PubMed: 14871829]

61. Tondera D, Grandemange S, Jourdain A, Karbowski M, Mattenberger Y, Herzig S, et al. SLP-2 is required for stress-induced mitochondrial hyperfusion. EMBO J. 2009; 28:1589–600. [PubMed: 19360003]

62. Bhandari P, Song M, Chen Y, Burelle Y, Dorn GW 2nd. Mitochondrial contagion induced by Parkin deficiency in Drosophila hearts and its containment by suppressing mitofusin. Circ Res. 2014; 114:257–65. [PubMed: 24192653]

63. Parone PA, Da Cruz S, Tondera D, Mattenberger Y, James DI, Maechler P, et al. Preventing mitochondrial fission impairs mitochondrial function and leads to loss of mitochondrial DNA. PLoS One. 2008; 3:e3257. [PubMed: 18806874]

64. Whelan RS, Konstantinidis K, Wei AC, Chen Y, Reyna DE, Jha S, et al. Bax regulates primary necrosis through mitochondrial dynamics. Proc Natl Acad Sci U S A. 2012; 109:6566–71. [PubMed: 22493254]

65. Rambold AS, Kostelecky B, Lippincott-Schwartz J. Together we are stronger: fusion protects mitochondria from autophagosomal degradation. Autophagy. 2011; 7:1568–9. [PubMed: 22024745]

66. Lee Y, Lee HY, Hanna RA, Gustafsson AB. Mitochondrial autophagy by Bnip3 involves Drp1-mediated mitochondrial fission and recruitment of Parkin in cardiac myocytes. Am J Physiol Heart Circ Physiol. 2011; 301:H1924–31. [PubMed: 21890690]

67. Youle RJ, Narendra DP. Mechanisms of mitophagy. Nat Rev Mol Cell Biol. 2011; 12:9–14. [PubMed: 21179058]

68. Jin SM, Lazarou M, Wang C, Kane LA, Narendra DP, Youle RJ. Mitochondrial membrane potential regulates PINK1 import and proteolytic destabilization by PARL. J Cell Biol. 2010; 191:933–42. [PubMed: 21115803]

69. Narendra D, Tanaka A, Suen DF, Youle RJ. Parkin is recruited selectively to impaired mitochondria and promotes their autophagy. J Cell Biol. 2008; 183:795–803. [PubMed: 19029340]

70. Okatsu K, Oka T, Iguchi M, Imamura K, Kosako H, Tani N, et al. PINK1 autophosphorylation upon membrane potential dissipation is essential for Parkin recruitment to damaged mitochondria. Nature communications. 2012; 3:1016.

71. Yang JY, Yang WY. Bit-by-bit autophagic removal of parkin-labelled mitochondria. Nature communications. 2013; 4:2428.

72. Kubli DA, Zhang X, Lee Y, Hanna RA, Quinsay MN, Nguyen CK, et al. Parkin protein deficiency exacerbates cardiac injury and reduces survival following myocardial infarction. J Biol Chem. 2013; 288:915–26. [PubMed: 23152496]

73. Billia F, Hauck L, Konecny F, Rao V, Shen J, Mak TW. PTEN-inducible kinase 1 (PINK1)/Park6 is indispensable for normal heart function. Proc Natl Acad Sci U S A. 2011; 108:9572–7. [PubMed: 21606348]

74. Siddall HK, Yellon DM, Ong SB, Mukherjee UA, Burke N, Hall AR, et al. Loss of PINK1 increases the heart's vulnerability to ischemia-reperfusion injury. PLoS One. 2013; 8:e62400. [PubMed: 23638067]

75. Quinsay MN, Thomas RL, Lee Y, Gustafsson AB. Bnip3-mediated mitochondrial autophagy is independent of the mitochondrial permeability transition pore. Autophagy. 2010; 6:855–62. [PubMed: 20668412]

76. Kubli DA, Ycaza JE, Gustafsson AB. Bnip3 mediates mitochondrial dysfunction and cell death through Bax and Bak. Biochem J. 2007; 405:407–15. [PubMed: 17447897]

77. Novak I, Kirkin V, McEwan DG, Zhang J, Wild P, Rozenknop A, et al. Nix is a selective autophagy receptor for mitochondrial clearance. EMBO Rep. 2010; 11:45–51. [PubMed: 20010802]

78. Hamacher-Brady A, Brady NR, Logue SE, Sayen MR, Jinno M, Kirshenbaum LA, et al. Response to myocardial ischemia/reperfusion injury involves Bnip3 and autophagy. Cell Death Differ. 2006; 14:146–57. [PubMed: 16645637]

79. Hanna RA, Quinsay MN, Orogo AM, Giang K, Rikka S, Gustafsson AB. Microtubule-associated protein 1 light chain 3 (LC3) interacts with Bnip3 protein to selectively remove endoplasmic reticulum and mitochondria via autophagy. J Biol Chem. 2012; 287:19094–104. [PubMed: 22505714]

80. Dorn GW 2nd. Mitochondrial pruning by Nix and BNip3: an essential function for cardiac-expressed death factors. J Cardiovasc Transl Res. 2010; 3:374–83. [PubMed: 20559783]

81. Chu CT, Ji J, Dagda RK, Jiang JF, Tyurina YY, Kapralov AA, et al. Cardiolipin externalization to the outer mitochondrial membrane acts as an elimination signal for mitophagy in neuronal cells. Nat Cell Biol. 2013; 15:1197–205. [PubMed: 24036476]

82. Liu L, Feng D, Chen G, Chen M, Zheng Q, Song P, et al. Mitochondrial outer-membrane protein FUNDC1 mediates hypoxia-induced mitophagy in mammalian cells. Nat Cell Biol. 2012; 14:177–85. [PubMed: 22267086]

83. Schaper J, Froede R, Hein S, Buck A, Hashizume H, Speiser B, et al. Impairment of the myocardial ultrastructure and changes of the cytoskeleton in dilated cardiomyopathy. Circulation. 1991; 83:504–14. [PubMed: 1991369]

84. Chen L, Gong Q, Stice JP, Knowlton AA. Mitochondrial OPA1, apoptosis, and heart failure. Cardiovasc Res. 2009; 84:91–9. [PubMed: 19493956]

85. Chen Y, Liu Y, Dorn GW 2nd. Mitochondrial fusion is essential for organelle function and cardiac homeostasis. Circ Res. 2011; 109:1327–31. [PubMed: 22052916]

86. Papanicolaou KN, Kikuchi R, Ngoh GA, Coughlan KA, Dominguez I, Stanley WC, et al. Mitofusins 1 and 2 are essential for postnatal metabolic remodeling in heart. Circ Res. 2012; 111:1012–26. [PubMed: 22904094]

87. Hom J, Yu T, Yoon Y, Porter G, Sheu SS. Regulation of mitochondrial fission by intracellular Ca2+ in rat ventricular myocytes. Biochim Biophys Acta. 2010; 1797:913–21. [PubMed: 20347716]

88. Ashrafian H, Docherty L, Leo V, Towlson C, Neilan M, Steeples V, et al. A mutation in the mitochondrial fission gene Dnm1l leads to cardiomyopathy. PLoS Genet. 2010; 6:e1001000. [PubMed: 20585624]

89. Sciarretta S, Zhai P, Shao D, Zablocki D, Nagarajan N, Terada LS, et al. Activation of Nox4 in the Endoplasmic Reticulum Promotes Cardiomyocyte Autophagy and Survival During Energy Stress Through the PERK/eIF-2alpha/ATF4 Pathway. Circ Res. 2013 in press.

90. Ong SB, Subrayan S, Lim SY, Yellon DM, Davidson SM, Hausenloy DJ. Inhibiting mitochondrial fission protects the heart against ischemia/reperfusion injury. Circulation. 2010; 121:2012–22. [PubMed: 20421521]

91. Javadov S, Rajapurohitam V, Kilic A, Hunter JC, Zeidan A, Said Faruq N, et al. Expression of mitochondrial fusion-fission proteins during post-infarction remodeling: the effect of NHE-1 inhibition. Basic Res Cardiol. 2011; 106:99–109. [PubMed: 20886221]

92. Shenouda SM, Widlansky ME, Chen K, Xu G, Holbrook M, Tabit CE, et al. Altered mitochondrial dynamics contributes to endothelial dysfunction in diabetes mellitus. Circulation. 2011; 124:444–53. [PubMed: 21747057]

93. Marsboom G, Toth PT, Ryan JJ, Hong Z, Wu X, Fang YH, et al. Dynamin-related protein 1-mediated mitochondrial mitotic fission permits hyperproliferation of vascular smooth muscle cells and offers a novel therapeutic target in pulmonary hypertension. Circ Res. 2012; 110:1484–97. [PubMed: 22511751]

94. Ishihara N, Nomura M, Jofuku A, Kato H, Suzuki SO, Masuda K, et al. Mitochondrial fission factor Drp1 is essential for embryonic development and synapse formation in mice. Nat Cell Biol. 2009; 11:958–66. [PubMed: 19578372]

95. Wakabayashi J, Zhang Z, Wakabayashi N, Tamura Y, Fukaya M, Kensler TW, et al. The dynamin-related GTPase Drp1 is required for embryonic and brain development in mice. J Cell Biol. 2009; 186:805–16. [PubMed: 19752021]

96. Wang X, Su B, Fujioka H, Zhu X. Dynamin-like protein 1 reduction underlies mitochondrial morphology and distribution abnormalities in fibroblasts from sporadic Alzheimer's disease patients. Am J Pathol. 2008; 173:470–82. [PubMed: 18599615]

97. Cassidy-Stone A, Chipuk JE, Ingerman E, Song C, Yoo C, Kuwana T, et al. Chemical inhibition of the mitochondrial division dynamin reveals its role in Bax/Bak-dependent mitochondrial outer membrane permeabilization. Dev Cell. 2008; 14:193–204. [PubMed: 18267088]

98. So EC, Hsing CH, Liang CH, Wu SN. The actions of mdivi-1, an inhibitor of mitochondrial fission, on rapidly activating delayed-rectifier K(+) current and membrane potential in HL-1 murine atrial cardiomyocytes. Eur J Pharmacol. 2012; 683:1–9. [PubMed: 22374256]

99. Gharanei M, Hussain A, Janneh O, Maddock H. Attenuation of doxorubicin-induced cardiotoxicity by mdivi-1: a mitochondrial division/mitophagy inhibitor. PLoS One. 2013; 8:e77713. [PubMed: 24147064]

100. Sharp WW, Fang YH, Han M, Zhang HJ, Hong Z, Banathy A, et al. Dynamin-related protein 1 (Drp1)-mediated diastolic dysfunction in myocardial ischemia-reperfusion injury: therapeutic benefits of Drp1 inhibition to reduce mitochondrial fission. FASEB J. 2014; 28:316–26. [PubMed: 24076965]

101. Disatnik MH, Ferreira JC, Campos JC, Gomes KS, Dourado PM, Qi X, et al. Acute inhibition of excessive mitochondrial fission after myocardial infarction prevents long-term cardiac dysfunction. J Am Heart Assoc. 2013; 2:e000461. [PubMed: 24103571]

102. Gao D, Zhang L, Dhillon R, Hong TT, Shaw RM, Zhu J. Dynasore protects mitochondria and improves cardiac lusitropy in Langendorff perfused mouse heart. PLoS One. 2013; 8:e60967. [PubMed: 23596510]

103. Givvimani S, Munjal C, Tyagi N, Sen U, Metreveli N, Tyagi SC. Mitochondrial division/mitophagy inhibitor (Mdivi) ameliorates pressure overload induced heart failure. PLoS One. 2012; 7:e32388. [PubMed: 22479323]

104. Chang YW, Chang YT, Wang Q, Lin JJ, Chen YJ, Chen CC. Quantitative phosphoproteomic study of pressure-overloaded mouse heart reveals dynamin-related protein 1 as a modulator of cardiac hypertrophy. Mol Cell Proteomics. 2013; 12:3094–107. [PubMed: 23882026]

105. Hong Z, Kutty S, Toth PT, Marsboom G, Hammel JM, Chamberlain C, et al. Role of dynamin-related protein 1 (Drp1)-mediated mitochondrial fission in oxygen sensing and constriction of the ductus arteriosus. Circ Res. 2013; 112:802–15. [PubMed: 23334860]

Highlights

- Fission and fusion are essential for maintaining the quality of mitochondria.
- Drp1 translocates to damaged mitochondria and induces fission.
- Dysregulation of fission/fusion leads to mitochondrial dysfunction and cell death.

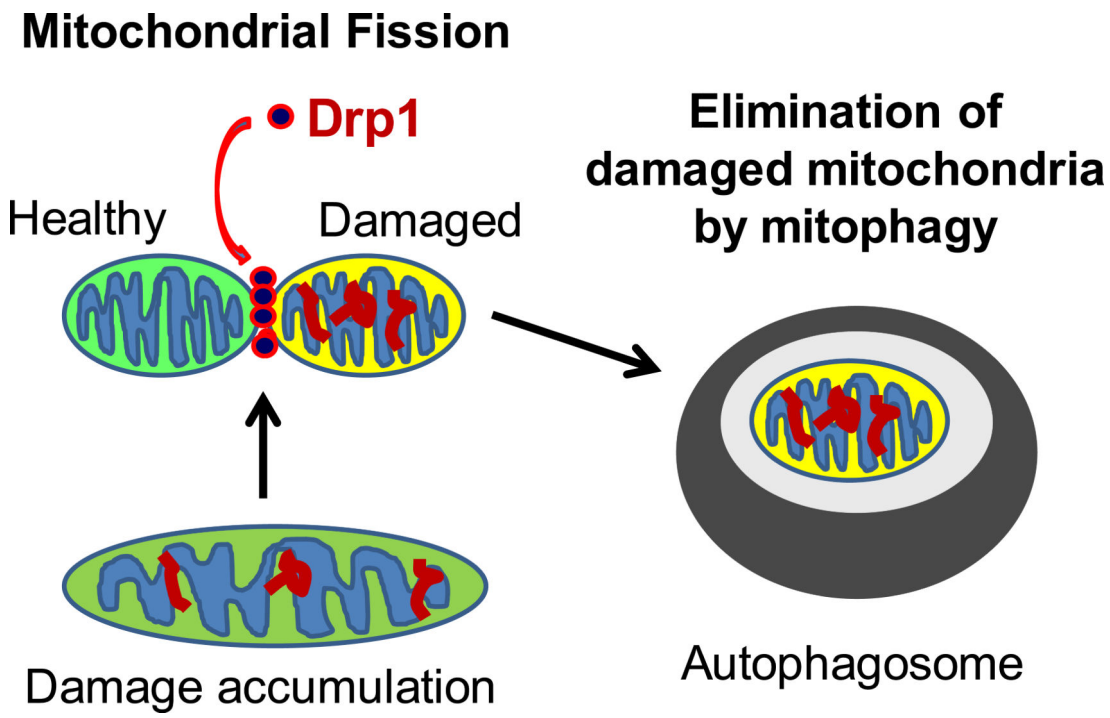

Figure 1. Elimination of damaged mitochondria by mitochondria-targeted selective autophagy, "mitophagy"

Mitochondrial health is maintained through mitochondrial dynamics (fission and fusion) and mitophagy. If damage accumulates in mitochondria, the mitocho ndria are aggregated and segregated by fission. This is followed by elimination of the damaged mitochondria via the autophagic process known as mitophagy. Drp1 plays an essential role in mediating mitochondrial fission.

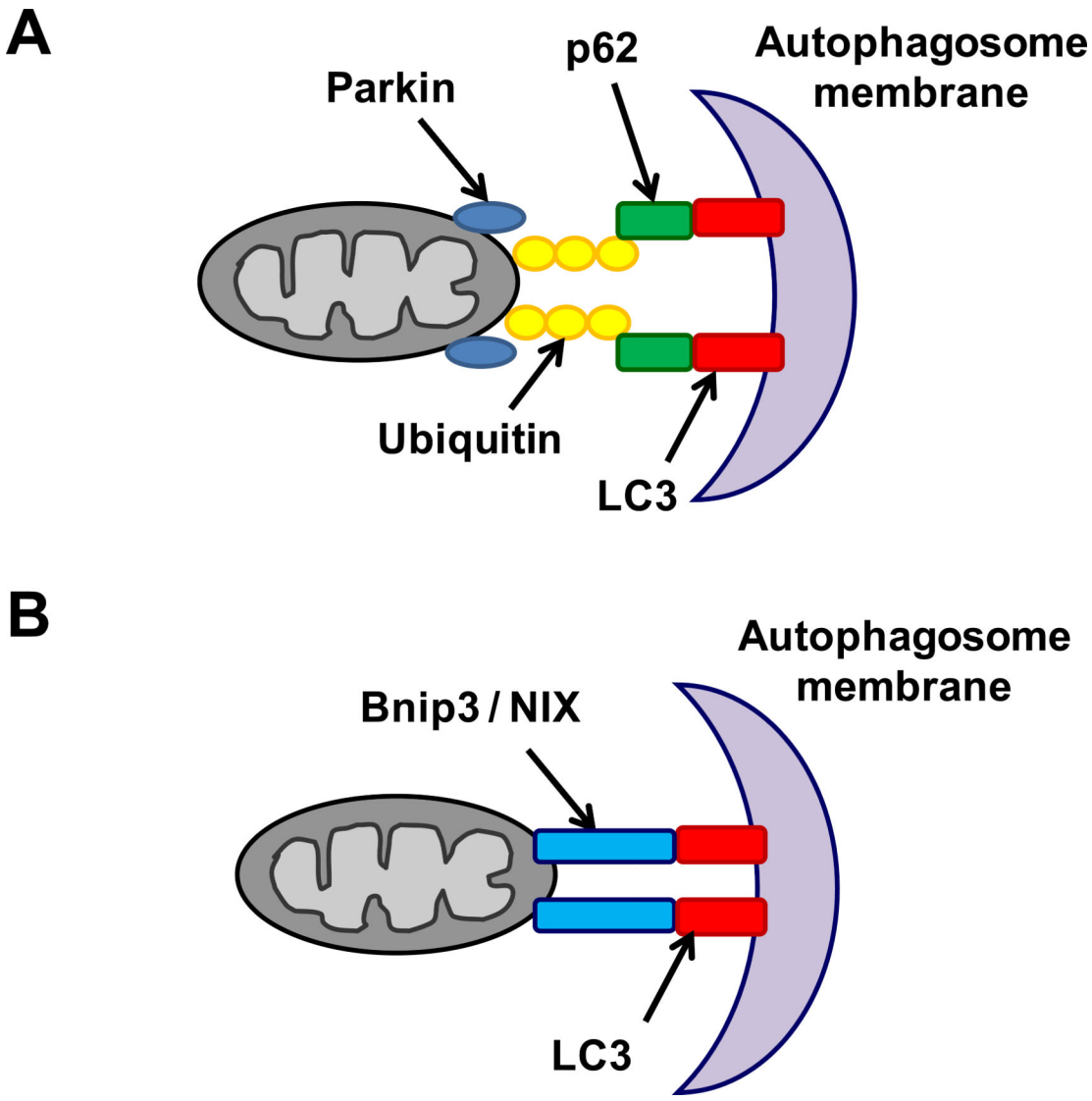

Figure 2. Mechanisms of mitophagic machinery

A The mitochondrially targeted serine/threonine kinase, PINK1, is expressed and de graded in healthy mitochondria. Upon membrane depolarization, PINK1 is stabilized and accumulates on the mitochondrial outer membrane, which leads to translocation of the cytosolic E3-ubiquitin ligase, Parkin, to mitochondria. Parkin ubiquitinates depolari zed mitochondria, leading to interaction with ubiquitin-binding deacetylase HDAC6 and p62/SQSTM1, which connects with LC3 and promotes assembly of the autophagic machinery. **B** Bnip3 and NIX are pro-apoptotic BH3-only proteins that facilitate opening of the mitochondrial permeability transition pore and activate Bax/Bak to permeabilize the mitochondrial membrane. Besides their pro-apo p totic effect, Bnip3 and NIX interact directly with LC3 and GABARAP on the phagophore, serving as a receptor for autophagosomes and tethering them to mitochondria. Recent evidence suggests that both FUNDC1 and cardiolipin also serve as receptors for LC3 and are involved in autophagy.

Table 1

The role of Drp1 in cardiovascular cells

Models	Species	Fission	Phenotype	Drp1 inhibitor	Reference
Dilated cardiomyopathy	Human	Enhanced	LV dysfunction	-	[83]
Post myocardial infarction	Rat	Enhanced	LV dysfunction	-	[84]
Cardiac specific Mfn1 and Mfn2 KO	Mouse	Enhanced	LV dysfunction	-	[85, 86]
Ceramide-induced apoptosis in CMs	Rat	Enhanced	Apoptosis	-	[33]
Doxorubicin-induced cardiomyopathy	Rat	Enhanced	Apoptosis	-	[33]
Ca2+ - mediated ROS generation in CMs	Rat	Enhanced	ROS generation	-	[87]
Cardiac hypertrophy induced by alpha-agonist stimulation	Rat	Enhanced	Hypertrophy	-	[91]
Aortic endothelial cells cultured with high glucose	Human	Enhanced	EC dysfunction	-	[92]
Inhibition of Drp1 in the heart with I/R	Mouse	Suppressed	Decreased I/R injury	mdivi-1	[90]
Inhibition of Drp1 in the heart with doxorubicin and I/R (Ex vivo)	Rat	Suppressed	Decreased I/R injury	mdivi-1	[99]
Inhibition of Drp1 in the heart with I/R	Rat	Suppressed	Decreased I/R injury	mdivi-1	[100]
Inhibition of Drp1 in the heart with I/R	Rat	Suppressed	Decreased I/R injury and improved LV function	P110	[101]
Inhibition of Drp1 in the heart with I/R (Ex vivo)	Mouse	Suppressed	Decreased I/R injury	Dynasore	[102]
Inhibition of Drp1 in the heart with PO	Mouse	Suppressed	Improved LV function	mdivi-1	[103]
Inhibition of Drp1 in CMs with phenylephrine treatment	Rat	?	Suppression of hypertrophy	mdivi-1	[104]
Inhibition of Drp1 in the heart with PO	Mouse	?	Suppression of hypertrophy	mdivi-1	[104]
Inhibition of Drp1 in DA	Rabbit	?	Rescued DA closure	mdivi-1	[105]
Pulmonary artery with PAH	Rat	Suppressed	Improved hemodynamics	mdivi-1	[93]
Heterozygous Drp1(C425F) mutation in Python mice	Mouse	?	LV dysfunction	-	[88]