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REVIEW

SUBJECT COLLECTION: MITOCHONDRIA

Mitochondria-associated niches in health and disease

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ABSTRACT

The appreciation of the importance of interorganelle contacts has steadily increased over the past decades. Advances in imaging, molecular biology and bioinformatic techniques allowed the discovery of new mechanisms involved in the interaction and communication between organelles, providing novel insights into the inner works of a cell. In this Review, with the mitochondria under the spotlight, we discuss the most recent findings on the mechanisms mediating the communication between organelles, focusing on Ca^{2+} signaling, lipid exchange, cell death and stress responses. Notably, we introduce a new integrative perspective to signaling networks that is regulated by interorganelle interactions – the mitochondria-associated niches – focusing on the link between the molecular determinants of contact sites and their functional outputs, rather than simply physical and structural communication. In addition, we highlight the neuropathological and metabolic implications of alterations in mitochondria-associated niches and outline how this concept might improve our understanding of multi-organelle interactions.

KEY WORDS: Mitochondria, Stress responses, Apoptosis, Mitochondria-associated membranes, MAMs, Bioenergetics

Introduction

The endosymbiotic theory, first formulated more than a hundred years ago, posits the origin of mitochondria as descendants of free-living prokaryotes that developed a symbiotic relationship with primitive host cells (Gray et al., 1999). This theory has opened new avenues for our understanding of cells and the evolution of eukaryotic organisms (Lazcano and Peretó, 2017). The term mitochondria-associated membranes (MAMs) was coined after the initial discovery that the endoplasmic reticulum (ER) has a bona fide interaction domain with mitochondria, which was required for proper lipid synthesis (Vance, 1990). In the past decade, accumulating studies have defined MAMs as sites of physical communication between mitochondria and the ER, where molecular exchanges are facilitated through the mediation of tethering molecules (Prudent and McBride, 2017). Moreover, another term to describe these regions of membrane physical closeness is also used in the field; ER–mitochondria contact sites (ERMCSs). With recent advances in high-resolution live-cell microscopy (Sigal et al., 2018; Wu et al., 2021), as well as the implementation of machine-learning algorithms in microscopy (Chen et al., 2021), new mechanisms and interactions have come to light that have helped researchers to better understand the internal organization of the cell and its organelles and metabolic pathways, based on a dynamic view of the intracellular network.

In this Review, we highlight the importance of the interconnectivity between mitochondria and other organelles for overall cell function, and contextualize the underlying mechanisms, which include regulation of membrane dynamics, Ca^{2+} signaling and lipid exchange in both health and disease. In addition, we explore interorganelle responses to stress and how an imbalance in these responses promotes disease development and outline future prospects for therapeutic intervention.

Importantly, we propose here the concept of mitochondria-associated niches (MANs) as a function-oriented term. We believe that the term MAMs does not capture the full scope of organelle interactions, as it is conceptually tied primarily to structural interactions with and/or juxtaposition of mitochondrial membranes. As the highly dynamic nature of these interactions is now well defined, we propose that the concept of MANs might better evoke the true meaning of these intracellular niches between different organelles that together control a large number of highly regulated cellular processes, as detailed herein.

Mitochondria-associated membrane dynamics

The processes by which mitochondria change their shape and overall structure are known as mitochondrial membrane dynamics and comprise regulated mitochondrial fission and fusion. The balance between fission and fusion contributes to mitochondrial quality control and responds to various metabolic pathways, such as autophagy, cell death and bioenergetics (Herrera-Cruz and Simmen, 2017; Prudent and McBride, 2017), that are closely linked to the maintenance of health or the occurrence of disease (Yapa et al., 2021). Given the independent evolutionary origin of mitochondria, it was initially thought that the dynamics of mitochondrial membranes depended solely on the mitochondria themselves (Chan, 2020), but ample evidence now shows that other organelles, notably the ER, also play a crucial role.

Fission

Mammalian mitochondrial fission is responsible for the generation of new mitochondria and is regulated by several processes, such as cell division, mitochondrial localization, mitophagy, bioenergetics and multiple stress pathways (Chan, 2020). The mitochondrial fission machinery has also been extensively studied as an important regulator in other highly coordinated processes, such as cell death (detailed in Box 1).

However, there are aspects of mitochondrial fission that are still not well understood. ERMCSs have been suggested as the site of mitochondrial constriction and fission, providing a conduit for the transport of Ca^{2+} and lipids (Friedman et al., 2011; Kornmann et al., 2009; Rizzuto et al., 1998; Wu et al., 2018). Recent studies have shown that mitochondrial fission occurs at particular sites where the ER contacts the mitochondria, more specifically in mitochondrial DNA (mtDNA)-rich regions (Lewis et al., 2016). At these sites, ER tubules wrap around the mitochondria to create an initial constriction that

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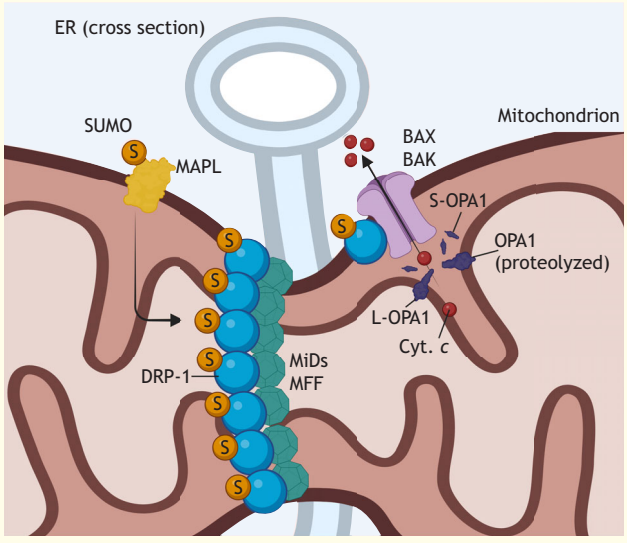
Box 1. The mitochondrial fission–apoptosis niche

Apoptosis is a highly coordinated process that controls cell fate in order to maintain tissue homeostasis. The main pathway that regulates apoptosis assembles at the mitochondrial and ER levels, and is orchestrated by pro- and anti-apoptotic proteins of the BCL-2 family (Youle and Strasser, 2008). Ultimately, antagonism between these proteins regulates the permeabilization of the OMM and the release of cytochrome *c* and other molecules into the cytosol via a pore formed by the oligomerization of Bcl-2-associated X protein (BAX) and Bcl-2 homologous antagonist/killer (BAK) (Youle and Strasser, 2008). DRP-1 has been shown to be required for cytochrome *c* release and apoptosis (Frank et al., 2001), although it was later found not to be required for pronounced mitochondrial structural changes at the onset of apoptosis (Milani et al., 2019). Nevertheless, DRP-1 temporarily associates with BAX during apoptosis, as BAX foci have been found in regions of mitochondrial fission during cell death (Karbowski et al., 2002). Furthermore, DRP-1-mediated mitochondrial membrane remodeling has been shown to facilitate BAX oligomerization during apoptosis (Montessuit et al., 2010), and, more recently, DRP-1 and BAX were observed to directly interact and induce apoptosis (Jenner et al., 2022) (see figure). DRP-1 is also essential for BH3 mimetic-induced apoptosis (Milani et al., 2017) and for remodeling of the mitochondrial cristae during apoptosis, the latter by facilitating OPA1 proteolysis, a process previously thought to be independent of the mitochondrial fission machinery (Otera et al., 2016). It is known that posttranslational modifications of DRP-1, in particular phosphorylation, regulate its fission activity (van der Bliek et al., 2013). In addition, DRP-1 SUMOylation (marked by S in the figure) has been shown to directly induce apoptosis. Mitochondrial ubiquitin ligase activator of NFκB 1 (MAPL; also known as MUL1), a mitochondrial-anchored RING-finger-containing protein, SUMOylates DRP-1 in response to multiple apoptotic stimuli and is essential for cytochrome *c* release downstream of BAX–BAK oligomerization at the OMM (Prudent et al., 2015) (see figure). Interestingly, the same study showed that MAPL stabilizes ER–mitochondria junctions and that this is required for proper Ca^{2+} exchange during apoptosis. Taken together, these studies strongly suggest that DRP-1 is not only the master regulator of mitochondrial fission but is also indispensable for the proper induction of cell death by bridging two tightly regulated processes, mitochondrial fission and apoptosis. Furthermore, as a fission protein that constricts mitochondria, DRP-1 physically promotes the profound mitochondrial membrane and shape alterations changes that are now beginning to be considered as an integral part of the apoptotic process (Prudent et al., 2015).



promotes recruitment of GTPase dynamin-related protein 1 (DRP-1; also known as DNM1L) to its receptors, mitochondrial fission factor (MFF), mitochondrial dynamics protein MID49 (MiD-49; also known as MIEF2) and the mitochondrial dynamics protein MID51 (MiD-51; also known as MIEF1) in the outer mitochondrial membrane (OMM) (Palmer et al., 2013), followed by DRP-1 oligomerization and the assembly of a constriction ring that leads to its scission (Macdonald et al., 2014). Recent extensions of this model further include the ER, components of the cytoskeleton, lysosomes and mitochondria. The regulated integration and coordination of these components make mitochondrial fission a *de facto* MAN (Fig. 1). Here, the initial tether, comprising inverted formin-2 (INF2) in the ER and Spire1C (also known as SPIRE1) in the OMM, acts as a scaffold for the assembly of actin and myosin IIA filaments, generating a mechanical force that compresses the mitochondrial membrane and initiates the recruitment of DRP-1 (Korobova et al., 2013, 2014; Manor et al., 2015).

In addition, lysosomes have recently been identified as key players in mitochondrial fission. The master regulator of lysosomal dynamics, GTP-bound Ras-related protein Rab7 (herein referring to both Rab7a and Rab7b forms), mediates the tethering of lysosomes and mitochondria, which is counteracted by mitochondrial fission 1 protein (Fis1)-mediated activation of TBC1D15 (a Rab7 GTPase-activating protein) (Onoue et al., 2013). At these contact points, Fis1 also regulates the fission of peripherally damaged mitochondria in an ER-independent manner, suggesting that processes such as mitochondrial fission can be differentially regulated by different organelles (Kleele et al., 2021) (Fig. 1). Moreover, in some cases of Parkinson's disease (PD), inhibition of TBC1D15 by a mutant form of the lysosomal enzyme β-glucocerebrosidase (GBA1) prolonged lysosomal–mitochondrial tethering and disrupted mitochondrial function (Kim et al., 2021b). Additionally, the Ca^{2+}-dependent phospholipid-interacting protein annexin A6 (AnxA6), found in endolysosomal membranes, has been shown to promote TBC1D15-mediated Rab7 inactivation (Meneses-Salas et al., 2020) and DRP-1 inhibition in the absence of Ca^{2+} (Chlystun et al., 2013), further underscoring the multi-organelle interconnectivity of the mitochondrial fission niche.

The final steps of mitochondrial fission are not yet well understood. Some studies suggest a role for dynamin 2 (Dyn2) in the final scission of mitochondria (Lee et al., 2016), although this is not fully required (Fonseca et al., 2019; Kamerkar et al., 2018). Other work suggests an involvement of phosphatidylinositol 4-phosphate (PI4P)-containing vesicles from the trans-Golgi network (TGN), as PI4P might be required for mitochondrial cleavage downstream of DRP-1 (Hung et al., 2017). ADP-ribosylation factor-1 (Arf1) or phosphatidylinositol 4-kinase IIIβ (PI4KIIIβ) have been associated with increased mitochondrial elongation and interconnectivity (Nagashima et al., 2020) (Fig. 1).

The interaction of mitochondria with other organelles, such as peroxisomes, endosomes, the plasma membrane and lipid droplets, can also affect the fission process. For example, the oxysterol-binding proteins (OSBP)-related proteins ORP5 and ORP8, known to transport phosphatidylserine (PS) from the ER to the plasma membrane, have been shown to be critical in the maintenance of mitochondrial morphology, cristae structure and respiratory activity (Tábara et al., 2021). In addition, the mitochondrial fission machinery also shares some components with other organelles; for example, peroxisomal division is mediated by DRP-1 and its adaptors MFF and Fis1 (Schrader et al., 2016), amassing evidence for the complexity and interconnectivity of multi-organelle membrane dynamics in this MAN.

Fusion

Mitochondrial fusion increases the overall mitochondrial capacity for oxidative phosphorylation and enables redistribution of

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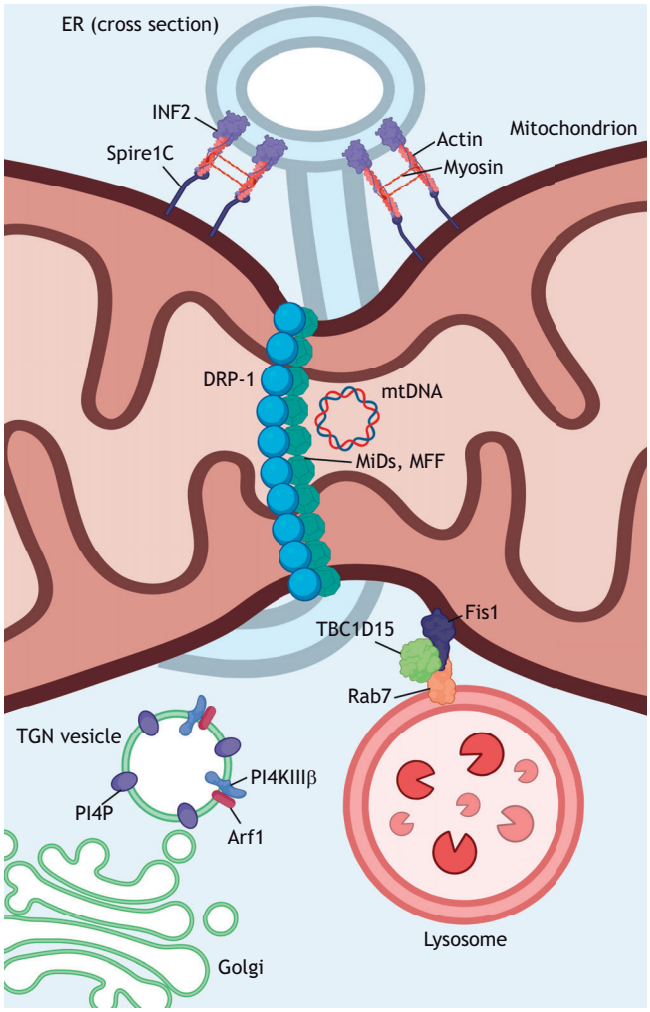

Fig. 1. The mitochondrial fission niche. Mitochondrial fission is finely regulated by the involvement of multiple cellular compartments. The fission process starts with mitochondrial DNA (mtDNA; middle) marking the site for fission. Then, the ER is recruited to the fission site by tethering INF2 to Spire1C in the mitochondria. Actin and myosin filaments allow the two membranes to come together and act to tighten the tether. Next, assembly of an oligomeric DRP-1 ring is facilitated by interaction with its adapters MiD49, MiD51 and MFF at the mitochondrial membrane, resulting in the scission of one mitochondrion into two. The late steps of mitochondrial fission are believed to require TGN vesicles containing PI4P, PI4KIIIβ and Arf1, given that the absence of one or more components promotes elongation. Mitochondrial–lysosomal physical interactions are also required for fission via TBC1D15-mediated Fis1 and Rab7 recruitment. Figure created with BioRender.com.

mtDNA in response to specific cellular energy demands and stress (Gao and Hu, 2021). In contrast to mitochondrial fission, the fusion machinery appears to be more confined to the mitochondria and less dependent on physical contact with the ER. When two mitochondria come closer together, in a movement mediated by the depolymerisation of actin filaments from the cytoskeletal network (Mattenberger et al., 2003), the GTPases mitofusin 1 and 2 (MFN1 and MFN2) join two adjacent OMMs, forming homotypic and heterotypic complexes (Hoppins et al., 2011). Fusion of inner mitochondrial membranes (IMMs) is mediated by optic atrophy 1 (OPA1) and is dependent on mitochondrial membrane polarization, as the stability of OPA1 is very sensitive to changes in the structural integrity of the IMM (Del Dotto et al., 2018). Furthermore, important mitochondrial fusion factors, such as MFN2, play a fundamental role in the structural connection between the ER and mitochondria (Naon et al., 2016). Similarly, the mitochondria and the ER-resident protein trichoplein (also known as myostatin), promote mitochondrial fusion by tethering ER and mitochondria via MFN2 (Cerqua et al., 2010), suggesting that mitochondrial fusion dynamics might also occur via MANs.

Molecular interplay of MANs

The molecular interplay between the ER and mitochondria has been extensively studied and includes numerous signaling pathways such as Ca^{2+} signaling, lipid metabolism, bioenergetics, inflammation, autophagy and apoptosis (Herrera-Cruz and Simmen, 2017; Scorrano et al., 2019), which have been traditionally associated with ERMCSs. As previously mentioned, we believe that these regions are not just a mere juxtaposition of two or more membranes, but that they function as communication hubs, spatially arranged niches of great importance for cellular function, as highlighted below.

Physiological signaling and exchanges of molecules

Ca^{2+} signaling MANs

The role of the Ca^{2+} signaling at the MAN consists of crosstalk between the ER and mitochondria, which plays important roles in multiple cellular pathways. The ER is the main intracellular Ca^{2+} store (De La Fuente et al., 2013; Somlyo, 1984), distributing Ca^{2+} to the cytosol via inositol 1,4,5-triphosphate receptors (IP_3Rs) (Streb et al., 1983). Under normal conditions, Ca^{2+} is highly concentrated in the ER lumen by the activity of the (sarco)endoplasmic reticulum Ca^{2+}-ATPase (SERCA), which actively pumps Ca^{2+} from the cytosol into the ER. From the ER, Ca^{2+} is transported to the mitochondria through IP_3R channels, taken up to the intermembrane space by the voltage-dependent anion-selective channel protein (VDAC), at the OMM, and finally entering the mitochondrial matrix via the mitochondrial Ca^{2+} uniporter complex (MCU), at the IMM (Marchi and Pinton, 2014; Fig. 2A). In the mitochondria, Ca^{2+} is essential for bioenergetics as it regulates Ca^{2+}-sensitive dehydrogenases of the tricarboxylic acid (TCA) cycle, ATP synthase and the malate-aspartate shuttle (Giorgi et al., 2018). In addition, constitutive Ca^{2+} transfer through the IP_3Rs is necessary to maintain adequate levels of reduced mitochondrial nicotinamide adenine dinucleotide (NADH), which is essential for oxidative phosphorylation and ATP production (Cárdenas et al., 2010). Furthermore, mitochondrial rapid micro-oxidative bursts that emerge from transient depolarization can stimulate IP_3R-mediated Ca^{2+} release in the context of stress or the presence of some apoptotic stimuli (Booth et al., 2021), suggesting that the regulation of Ca^{2+} transfer between these two organelles is more refined and complex than previously anticipated. Compared with the ER, the mitochondrial Ca^{2+} concentration is quite low under normal conditions (Somlyo, 1984), but can be increased by various stress conditions. For example, changes in the expression of tumor suppressors and oncogenes can modulate mitochondrial Ca^{2+} concentration and trigger specific cellular responses, such as apoptosis and necroptosis (Faizan and Ahmad, 2021). In contrast to the OMM, the IMM is impermeable to Ca^{2+}; therefore, the import of Ca^{2+} into the matrix is mainly mediated by the MCU complex (Marchi and Pinton, 2014) (Fig. 2A). In addition to bona fide channels, several ER structural proteins play additional roles in the regulation of Ca^{2+} signaling. For example, phosphofurin acidic cluster sorting protein 2 (PACS2), originally described as an ER–mitochondria tether, also modulates Ca^{2+}

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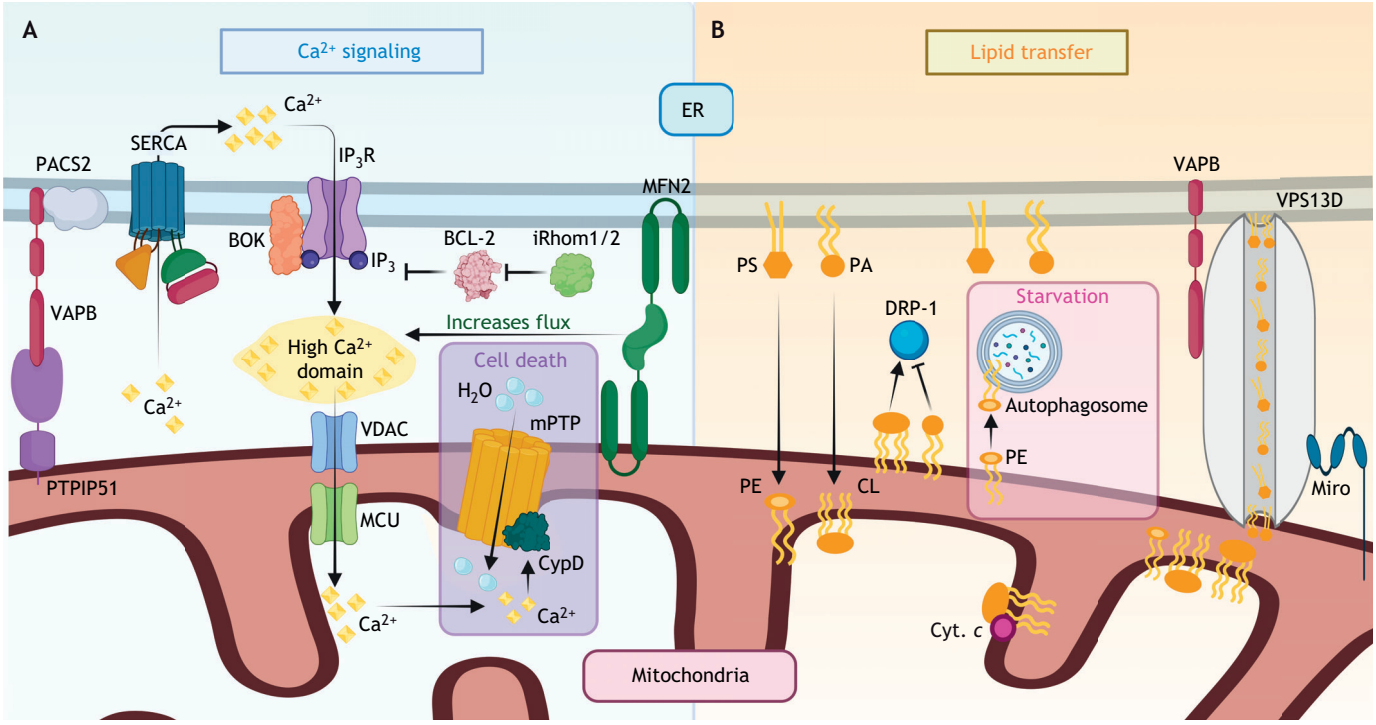

Fig. 2. Physiological MANs – Ca^{2+} signaling and lipid transfer. MANs orchestrate exchange of essential ions and macromolecules, ensuring proper cellular function. (A) Ca^{2+} signaling. PACS2-induced binding of VAPB to PTPIP51 facilitates Ca^{2+} exchange between the ER and mitochondria. SERCA is responsible for restoring ER Ca^{2+}. IP3R mediates the release of Ca^{2+} from the ER into the cytosol, creating a high-Ca^{2+} domain from which Ca^{2+} enters the mitochondria via VDAC and MCU. Mitochondrial Ca^{2+} increases cyclophilin D (CypD) activity, opening the mPTP and allowing H_2O influx, resulting in mitochondrial swelling and cell death. The antiapoptotic protein BCL-2 inhibits IP_3R activity and is inhibited by iRhom1 and iRhom2 (iRhom1/2). MFN2-mediated tethering between the ER and mitochondria can increase mitochondrial Ca^{2+} influx. (B) Lipid transfer. PS is synthesized in the ER and transferred to the mitochondria where it is converted into PE. Similarly, PA, an ER lipid, is converted to CL at the mitochondria. CL-rich membranes facilitate recruitment of DRP-1 to fission sites, whereas PA-rich regions inhibit it. Upon starvation, mitochondria can feed autophagosomes with lipids. VPS13D, together with VAPB and Miro, can facilitate lipid transfer from the ER to the mitochondria. Figure created with BioRender.com.

signaling (Simmen et al., 2005). The ER–mitochondrial tether MFN2 also enables Ca^{2+} signaling, allowing greater sensitivity to apoptosis by increasing Ca^{2+} influx into the mitochondria (Guo et al., 2007; Wang et al., 2015) (Fig. 2A). Interestingly, other stimuli, such as hydrogen peroxide and superoxide, can also trigger Ca^{2+} influx from the ER into the mitochondria. For instance, MCU activity can be stimulated by the accumulation of reactive oxygen species (ROS) on ERMCSs as a result of increased Ca^{2+} efflux from the ER into the mitochondria (Dong et al., 2017). Finally, lysosomal proteins, such as the transient receptor potential channel mucolipin 1 (TRPML1), have also been found to mediate Ca^{2+} signaling alongside the ER and mitochondria, configuring a possible tripartite organelle Ca^{2+} MAN (see Box 2).

Lipid transfer MANs

Lipid transfer configures an important role of the MAN as it is one of the main means of ER–mitochondrial crosstalk and one of the first functions reported for ERMCSs, originally identified as a PS-rich fraction (Vance, 2014). PS is synthesized in the ER and then transferred to the mitochondria where it is decarboxylated to phosphatidylethanolamine (PE) (Vance, 2008; Voelker, 1989) (Fig. 2B). Mitoguardin-2, an OMM protein, has been shown to operate in this niche by mediating the transference of PS from lipid droplets to the ER through an interaction with vesicle-associated membrane protein-associated protein B/C (VAPB) (Kim et al., 2022). In addition, enzymes involved in cholesterol and sphingolipid synthesis are also located at ERMCSs (Fujimoto et al., 2012; Vance, 2014). Similarly, the chaperone Sigma-1R (Sig-1R; also known as SIGMAR1), which is implicated in neurodegenerative diseases, such as Alzheimer's disease (AD) and amyotrophic lateral sclerosis (ALS), associates with simple sphingolipids, such as ceramides, at the ER to regulate Ca^{2+} release (Hayashi and Fujimoto, 2010). The synthesis of phosphatidic acid (PA) in the ER and its conversion to cardiolipin (CL) in the mitochondria is an excellent example of a lipid exchange MAN. Considered as the signature phospholipid of the mitochondria, CL makes up ∼20% of the total lipid of the IMM and is crucial for important mitochondrial processes, such as respiration, apoptosis, mitophagy and membrane dynamics (Claypool and Koehler, 2012; Herrera-Cruz and Simmen, 2017; Kagan et al., 2005). Interestingly, the accumulation of PA in the mitochondria inhibits DRP-1 activity and thus mitochondrial fission (Adachi et al., 2016), underscoring the importance of these phospholipids not only as structural molecules but also as key players in the regulation of various processes at MANs including lipid exchange, membrane dynamics and cell death (Kameoka et al., 2018). This interconnectivity is further supported by a study showing that, in mammals, Miro (also known as RHOT1), a Ca^{2+}-activated GTPase that regulates mitochondrial dynamics and motility via microtubule interactions (Nemani et al., 2018), recruits the lipid transport protein vacuolar protein sorting 13 homolog D (VPS13D) to the ER, creating a lipid conduit to the mitochondria (Guillén-Samander et al., 2021) (Fig. 2B). Although the origin for the concept of MAMs, due to a lack of sensitive tools, the study of the lipid transfer niches in mammals is only recently expanding with the

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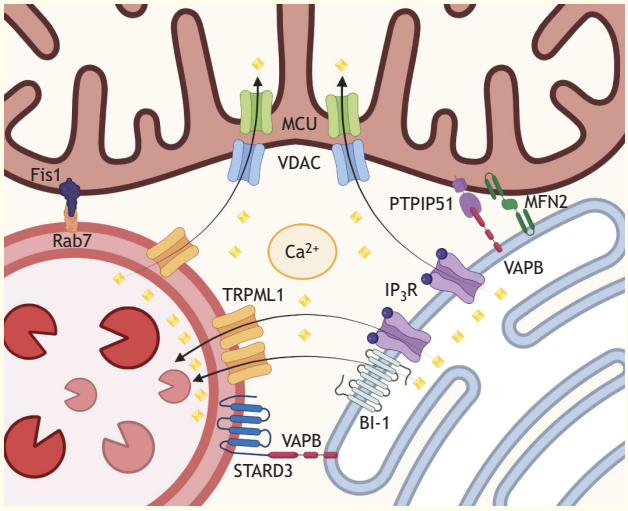
Box 2. The tripartite Ca^{2+} niche

Based on our concept of MANs, we propose here a tripartite niche that highlights the interactions between mitochondria, ER and lysosomes through Ca^{2+}, to regulate energy, metabolism, apoptosis and autophagy.

In this niche, Ca^{2+} is transferred from the lysosomes to the mitochondria via the transient receptor potential mucolipin 1 channel (TRPML1; also known as MCOLN1) in contact regions between these two organelles, mediated by tethers such as Fis1 and Rab7 (Onoue et al., 2013; Peng et al., 2020) (see figure). Ca^{2+} then enters the mitochondria via VDAC at the OMM and MCU at the IMM. In addition, alterations in the mitochondria–lysosomal contacts results in defective mitochondrial Ca^{2+} uptake from the lysosomes and underlies the lysosomal storage disorder mucolipidosis type IV, which is caused by mutations in TRPML1 (Peng et al., 2020). This is particularly interesting, as a previous study showed that the refilling of lysosomal Ca^{2+} occurs through the ER via IP_3Rs and not through the pH gradient as previously thought (Garrity et al., 2016), in a process facilitated by Bax inhibitor 1 (BI-1; also known as TMBIM6) at the ER (Kim et al., 2021a) and the tethers STARD3 and VAPB (Alpy et al., 2013). Therefore, a plausible Ca^{2+} exchange from the ER to lysosomes to mitochondria could drive multiple pathways involving these three organelles to maintain homeostasis, in addition to the well described IP_3R–VDAC–MCU complex, supported by additional tethers, such as MFN2, PTPIP51 and VAPB. Given that Ca^{2+} plays a crucial role in autophagy, it is not far fetched to speculate on a broader role for this niche in the process of providing a surface assembly for the autophagosome and a 2D surface for its fusion with the lysosome. The relevance of the tripartite niche becomes clearer when one considers the most recent model of mitochondrial fission, which includes these three organelles as regulators, as described in the main text and reviewed by the Prudent group (Tábara et al., 2021).

The mitochondrial–ER–lysosome Ca^{2+} exchange niche is a great example of how dynamic and complex the interactions between organelles are and how the entire intracellular system is orchestrated by its various parts. It further underscores the importance of looking at the cell in an integrative manner to deepen our understanding and generate new insights into health and disease.



support of new methods such as METALIC, a mass tag mass spectrometry-based technique for tracking specific lipids in the cells (John Peter et al., 2022).

Coping with stress

The unfolded protein response

In addition to forming niches of fission and molecular exchanges with the mitochondria, the ER is also responsible for the production of one-third of all the eukaryotic proteins for multiple cellular functions (Hetz et al., 2020). Different physiological and pathological conditions can alter the protein folding process at the ER and increase the level of misfolded proteins, leading to a condition known as ER stress. Cells cope with ER stress through mechanistic adaptations collectively named the unfolded protein response (UPR). In mammals, the UPR is orchestrated by three main signaling cascades that respond to the folding status of proteins in the ER lumen. The three arms of the UPR are initiated by three different ER transmembrane protein sensors: inositol-requiring enzyme 1 α (IRE1α), protein kinase RNA activated-like ER kinase (PERK; also known as EIF2AK3) and activating transcription factor 6 (ATF6) (Kaufman, 2002).

The PERK UPR arm was the first to be described as being regulated at the level of what we consider a MAN, based on studies describing morphological changes in mitochondria induced by PERK deficiency (Carreras-Sureda et al., 2019; Mori et al., 2013). During the UPR, PERK activation results in phosphorylation of the eukaryotic translation initiation factor 2 subunit-α (eIF2α), promoting transient inhibition of protein synthesis, while also inducing the translation of activating transcription factor 4 (ATF4), leading to increased expression of genes involved in stress responses (Kaufman, 2002; Ron and Walter, 2007) (Fig. 3A). Interestingly, PERK ablation has been shown to increase the number of contacts between ER and mitochondria and desensitize cells to ROS-induced cell death (Verfaillie et al., 2012). Furthermore, MFN2 has been shown to modulate the UPR by inhibiting PERK activity (Muñoz et al., 2013) and by controlling the kinetics of ER stress events, thereby preventing ER stress-induced apoptosis (Ngoh et al., 2012). PERK overexpression also induced mitochondrial fragmentation (Muñoz et al., 2013), highlighting the importance of the UPR at the interface of multiple cellular signaling pathways as well as mitochondrial membrane dynamics. PERK has been shown to modulate mitochondrial dynamics by promoting mitochondrial hyperfusion in the initial stages of ER stress, preventing pathological mitochondrial fragmentation (Lebeau et al., 2018). PERK can also increase oxidative phosphorylation (OXPHOS) by regulating the levels of the splicing factor arginine/serine-rich 19 (SCAF1) under conditions of nutrient starvation or ER stress (Balsa et al., 2019). Regulation of OXPHOS by PERK has been shown to promote import of the MICOS complex subunit MIC19 (MIC19; also known as CHCHD3) into the mitochondria, thereby inducing cristae formation upon occurrence of energy-intensive processes, such as cold stress or β-adrenergic stimuli (Latorre-Muro et al., 2021) (Fig. 3A). Similarly, a novel stress pathway termed the mitochondrial UPR (mtUPR), coordinated by OMA1–DELE1–HRI (HRI is also known as EIF2AK1) and induced by imbalances in mitochondrial proteostasis, is mediated by an integrated stress response that is relayed from the mitochondria (OMA1–DELE1) to the cytosol (DELE1–HRI), involving the activation of eIF2α and ATF4 in the cytosol (Fessler et al., 2020; Guo et al., 2020) (Fig. 3A). Collectively, these studies highlight how UPR mediators such as PERK and ATF4 can participate in multiple distinct MANs, having roles in adaptation to bioenergetic demands and stress responses that are not directly related to the canonical UPR, opening new avenues for further investigations on this topic.

The IRE1α arm of the UPR is activated by ER stress, which involves IRE1α autophosphorylation and activation of its RNase domain (Credle et al., 2005; Zhou et al., 2006). Upon activation, IRE1α catalyzes the splicing of *XBP1* into *XBP1s*, which leads to upregulation of genes involved in the translocation, folding and secretion of ER proteins (Calfon et al., 2002; Yoshida et al., 2001).

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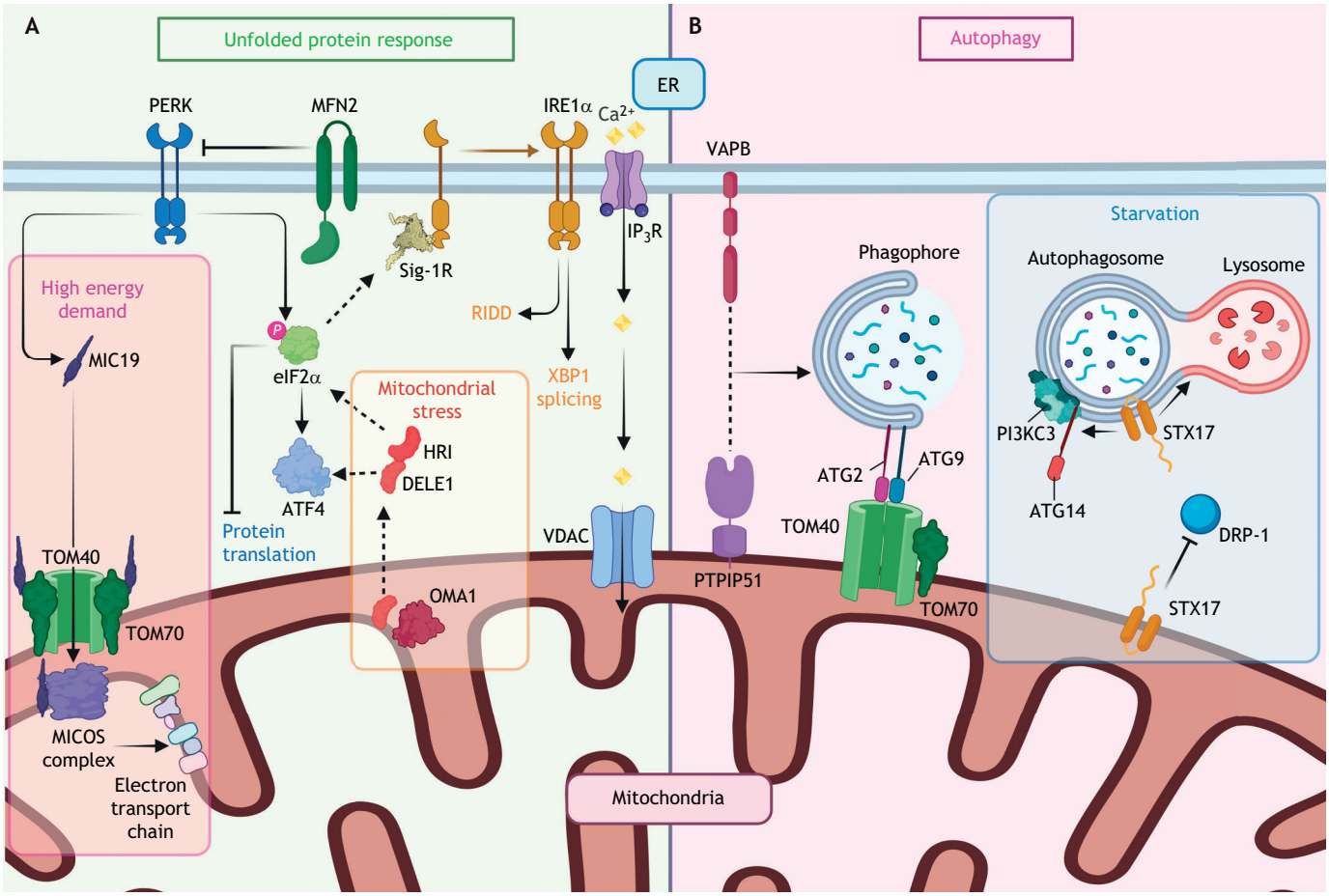

Fig. 3. Stress response MANs – UPR and autophagy. MANs organize crucial responses to stress to promote cell survival. (A) UPR signaling. PERK promotes phosphorylation of eIF2α, which inhibits protein translation and induces ATF4. Under high energy demand, PERK induces the TOM70-mediated translocation of the MIC19 subunit of the MICOS complex in mitochondria, thereby upregulating the electron transport chain. The mitochondrial UPR is mediated by the OMA1–DELE1–HRI pathway, which can induce eIF2α and ATF4 to cope with stress. Interaction with the chaperone Sig-1R promotes IRE1α oligomerization. IRE1α activity promotes splicing of XBP1 and RIDD, and acts as a scaffold for IP_3R-mediated Ca^{2+} release from the ER. (B) Autophagy. Tether proteins, such as VAPB and PTPIP51, facilitate the activation of autophagy in MANs. Autophagy is initiated by the formation of a phagophore, which matures into autophagosomes. Similarly, the mitochondrial complex TOM40–TOM70 interacts with ATG2 and ATG9 proteins to initiate phagophore formation and promote its expansion. Under starvation, STX17 dissociates from DRP-1 and mediates the translocation of ATG14 to the PI3KC3 complex on the autophagosome, promoting autophagy and inhibiting mitochondrial fission. In addition, in this context, STX17 promotes the fusion of autophagosomes with lysosomes during the final phase of autophagy. Figure created with BioRender.com.

In addition, IRE1α mediates the degradation of selected mRNAs and microRNAs through a process called regulated IRE1-dependent decay (RIDD), which is thought to mitigate ER protein overload by reducing RNA abundance (Hetz et al., 2020) (Fig. 3A). As previously mentioned, IRE1α is highly enriched in some MANs, where it plays an important role in several processes. Recently, our group described that IRE1α regulates Ca^{2+} signaling by acting as a scaffold for IP_3Rs, underscoring the role of IRE1α in mitochondrial bioenergetics at MANs. IRE1α ablation promotes mitochondrial metabolic stress by reducing ATP levels and activating the AMP-activated protein kinase (AMPK) pathway and autophagy (Carreras-Sureda et al., 2019). Sig-1R also brings together distinct niches as it is required for IRE1α-mediated responses to mitochondrial ROS and subsequent XBP1 mRNA splicing (Mori et al., 2013) (Fig. 3A), potentially coupling the UPR to lipid metabolism and linking two different aspects of MANs (Hayashi and Su, 2004).

Interestingly, the loss of MFN2 or PACS2 induces ER stress, while also untethering ER and mitochondria (Sebastián et al., 2012; Simmen et al., 2005). Furthermore, PACS2 has been found to be involved in ER stress by interacting with and retaining phosphorylated calnexin (CNX) on the ER (Myhill et al., 2008), thereby disrupting protein N-glycosylation and folding (Kozlov and Gehring, 2020).

Overall, the UPR requires a tight connection between the ER and the mitochondria to function properly, configuring a bona fide MAN. At the same time, proteins resident in these niches regulate the amplitude and duration of the UPR, thereby playing an important role in influencing cell fate.

Autophagy

Macroautophagy (hereafter autophagy) is an adaptive catabolic process by which intracellular macromolecules are broken down into simpler cellular elements such as amino acids, nucleotides and fatty acids, which are used by the cell to fuel metabolic pathways (Dikic and Elazar, 2018; Prinz et al., 2020). Autophagy is commonly initiated in response to stress conditions such as starvation, hypoxia and ER stress, and it is characterized by the formation and expansion of an isolation membrane named

phagophore into a closed double-membrane vesicle called an autophagosome in a sequential process coordinated by the members of the autophagy-related (ATG) proteins (Nishimura and Tooze, 2020) (Fig. 3B). During this process, the autophagosome sequesters various intracellular materials and later fuses with lysosomes to degrade intravesicular products (Levy et al., 2017). In mammals, autophagosome initiation and maturation occurs in close proximity to the so-called omegasome, a specialized subdomain of the ER enriched in phosphatidylinositol 3-phosphate (PI3P) (Hayashi-Nishino et al., 2009; Nishimura et al., 2017). In addition to the omegasome, the growing autophagosome simultaneously forms multiple contact sites with other organelles and membranes, including mitochondria, lysosomes, endosomes, the Golgi and the plasma membrane (Biazik et al., 2015). The role of the interactions between mitochondria and other organelles is of such importance for the execution of autophagy that we propose it configures an important MAN. For example, upon starvation, proteins of the Unc-51-like kinase-1 (ULK1) and the class III PI3K (PI3KC3) complexes, which normally diffuse freely in the ER membrane, relocate to contact sites (Hamasaki et al., 2013). Similarly, autophagy-related 14 (ATG14), Beclin 1, AMBRA1 and vacuolar protein sorting 34 (VPS34; also known as PIK3C3) – PI3KC3 complex components – assemble almost exclusively at this MAN to catalyze local PI3P production at the omegasome, thereby initiating the formation of the isolation membrane (Garofalo et al., 2016). Mitochondria are also involved in the formation of autophagosomes by transferring lipids directly from the OMM to the maturing autophagosome through a mechanism that requires interaction with the ER (Hailey et al., 2010). Furthermore, ATG2 and ATG9 proteins are recruited into mitochondria through interactions with the TOM40–TOM70 mitochondrial complex to induce phagophore maturation (Tang et al., 2019) (Fig. 3B). Interestingly, uncoupling the ER from the mitochondria by disrupting the interaction between the ER-resident protein VAPB and the OMM protein protein tyrosine phosphatase-interacting protein 51 (PTPIP51; also known as RMDN3) results in impaired Ca^{2+} transfer and the activation of survival autophagy (De Vos et al., 2012; Gomez-Suaga et al., 2017) (Fig. 3B). Therefore, proteins of this niche could have a dual, opposite effect on autophagy depending on the condition. On the one hand, this MAN promotes autophagy by providing a site for the assembly of the initiation complexes, and providing lipids and proteins for autophagosome elongation and maturation. On the other hand, a close and stable contact between the ER and the mitochondria is required for effective Ca^{2+} transfer, which maintains mitochondrial energy homeostasis.

Finally, there is a close interplay between mitochondrial dynamics and the induction of autophagy. Under nutrient-rich conditions, the autophagy protein STX17 associates with DRP-1 at sites of mitochondrial fission, where it promotes its activity by decreasing phosphorylation at S637, resulting in increased mitochondrial division. However, upon starvation, STX17 dissociates from DRP-1 and instead associates with ATG14, recruiting the autophagy PI3K3C initiation complex to this niche (Arasaki et al., 2015) (Fig. 3B). As a result, mitochondrial fission is inhibited during autophagy and mitochondrial elongation is favored. In addition, the ER could serve as a scaffold to facilitate autophagosome–lysosome fusion during the final stages of autophagy, as STX17 is required for fusion of the autophagosomes and lysosomes (Itakura et al., 2012). Given the complex interactivity of these processes, MANs act as signaling platforms, integrating information about the nutrient status of the cell to regulate autophagy as well as mitochondrial dynamics and metabolism.

Yielding to stress – cell death

Ca^{2+} is the major signaling metabolite involved in the regulation of cell death in MANs under stress conditions, and the ER is the major intracellular Ca^{2+} reservoir with an approximate intraluminal concentration ($[Ca^{2+}]_{ER}$) of 400–800 µM (Giorgi et al., 2018). As previously mentioned, mitochondria require a steady influx of Ca^{2+} to maintain their basal metabolism; however, excessive Ca^{2+} influx leads to overloading of the mitochondrial matrix, which increases the activity of cyclophilin D, followed by opening of the mitochondrial permeability transition pore (mPTP), resulting in mitochondrial swelling and the rupture of the inner and outer mitochondrial membranes (Baines et al., 2005) (Fig. 2A). Mitochondrial permeabilization results in the release of apoptogenic factors, including cytochrome *c*, and an overall breakdown in mitochondrial bioenergetics, leading to cell death (Green, 2019). Additionally, Ca^{2+} has been proposed to disintegrate the mitochondrial respiratory complex II by interfering with its binding to CL, leading to increased ROS production and cell death (Hwang et al., 2014).

The proteins of the B-cell lymphoma 2 (BCL-2) family are the best-studied accessory regulators of the IP_3Rs and Ca^{2+} signaling in the ER (Pihán et al., 2017). BCL-2 members are functionally divided into anti- and pro-apoptotic proteins, with the former inhibiting cell death by interacting with and sequestering proapoptotic proteins in the cytosol, whereas the latter promote mitochondrial outer membrane permeabilization (MOMP) and cell death in response to multiple cytotoxic stimuli including ER stress, DNA damage and viral infections (Kale et al., 2018; Singh et al., 2019). Typically, antiapoptotic BCL-2 family members, such as BCL-2 itself, BCL-X_L (encoded by *BCL2L1*), and MCL-1 bind to different regions of IP_3Rs on the ERMCS and inhibit Ca^{2+} release, mitochondrial uptake and cell death (Marchi et al., 2018b93; Prudent and McBride, 2017) (Fig. 2A). The inhibitory function of BCL-2 via IP_3R plays an important role in several pathophysiological processes and can be exploited to increase therapeutic efficacy in cancer (Akl et al., 2013; Bittremieux et al., 2019).

The antiapoptotic BCL-X_L has a similar function in regulating Ca^{2+} to BCL-2 and inhibits sustained Ca^{2+} leakage through IP_3R and cell death (Rosa et al., 2022). However, early studies revealed an additional function, where BCL-X_L interacts with the C-terminal domain of the IP_3R and sensitizes it to small concentrations of IP_3, resulting in the transmission of a small oscillating burst of Ca^{2+} to the mitochondria (Li et al., 2007; White et al., 2005). In the mitochondrial matrix, these oscillating Ca^{2+} bursts increase mitochondrial bioenergetics by making cells more resistant to proapoptotic stimuli (Yang et al., 2016). However, this model has been challenged by a recent study showing that BCL-X_L interacts with the same domains of IP_3R as BCL-2 always resulting in its inhibition, regardless of the concentration of IP_3 used (Rosa et al., 2022). Finally, several proteins can interact with and modulate the function of BCL-2 proteins, thereby indirectly regulating the activity of IP_3Rs. For example, the rhomboid-like superfamily pseudokinases iRhom1 and iRhom2 interact with and sequester BCL-2, releasing its inhibitory effects on the IP_3R (Fig. 2A), resulting in an increased Ca^{2+} release and mitochondrial uptake during ER stress (Dulloo et al., 2022). However, the precise contribution of this mechanism to ER stress-induced cell death has not yet been fully determined, as BCL-2 is also activated during ER stress by IRE1α and CHOP (also known as DDIT3), exerting additional regulatory functions in cell death beyond its role as an IP_3R inhibitor (Pihán et al., 2017).

The proapoptotic BCL-2 ovarian killer (BOK) also localizes in the ER, where it binds to the IP_3R and regulates its activity

(Echeverry et al., 2013; Schulman et al., 2013) (Fig. 2A). BOK is a less-studied member of the multidomain BCL-2 family and, unlike BAX and BAK (also known as BAK1), induces MOMP via a BCL-2-independent mechanism governed by its stability (Llambi et al., 2016). Interestingly, BOK has been shown to be required for the proper apposition and integrity of ERMCSs. Indeed, several ERMCS proteins, including the IP_3R, are missing in BOK-knockout cells, making the cell more resistant to stimulus-induced Ca^{2+} transfer and cell death. Furthermore, the correct localization of the IP_3R in ERMCS depends in part on its interaction with BOK, which is required for Ca^{2+} transfer to the mitochondria and apoptosis under ER stress (Carpio et al., 2021).

Members of the TMBIM family also control cell death at this niche. TMBIM6/BI-1 is located at MAMs and inhibits Ca^{2+} release from IP_3R at the ER membrane, preventing mitochondrial Ca^{2+} overload, opening of the mPTP and apoptosis (Rojas-Rivera and Hetz, 2015). By contrast, upon lysosomal perturbation, TMBIM1 promotes lysosomal BAX translocation to induce cell death via lysosomal membrane permeabilization (Pihán et al., 2021; Pihán et al., 2022).

Other molecular signaling pathways in MANs can also trigger cell death. For example, ceramide, an endogenous lipid mediator of apoptosis, triggers ER Ca^{2+} release and accumulation in the mitochondria, leading to mitochondrial fragmentation. BCL-2 and other antiapoptotic proteins inhibit the action of ceramide by lowering the intraluminal Ca^{2+} concentration in ER, thereby decreasing the pool of stimulus-dependent Ca^{2+} released (Pinton et al., 2001). Interestingly, some lysosomal storage diseases, such as GM1 gangliosidosis, which is caused by abnormal accumulation of the glycosphingolipid GM1 ganglioside, show accumulation of ceramide in this niche, where it binds directly to and regulates the IP_3R, increasing ER-to-mitochondria Ca^{2+} transfer and apoptosis (Sano et al., 2009). Further studies are needed to uncover the role of sphingolipids in MANs in regulating mitochondrial cell death.

Finally, PERK promotes the effective transfer of ROS from the ER to the mitochondria under ER stress induced by oxidation. As mentioned above, PERK acts as a tether, increasing both the number and proximity of ERMCSs, leading to the spread of ROS into the mitochondria with subsequent induction of cell death (Verfaillie et al., 2012).

Taken together, these studies illustrate that MANs act as complex hubs for both the regulation of mitochondrial bioenergetics and autophagy through Ca^{2+} signaling, and as sites for the induction of cell death under stress conditions. Ultimately, the outcome for the cell depends on the presence or absence of, or interactions with, various regulatory factors, including members of the BCL-2 protein family, IP_3Rs and components of the UPR, and the number and proximity of ER–mitochondrial junctions. Their presence establishes different signaling niches that strongly influence cell fate.

MANs in pathology

Dysfunctions of MANs have been implicated in the pathology of several metabolic and neurodegenerative diseases (further expanded in Table S1).

In PD, the functionality of the mitochondrial fission niche is altered by mutations in phosphatase and tensin homolog (PTEN)-induced kinase 1 (PINK1), which promotes DRP-1-mediated mitochondrial fragmentation and reduces mitochondrial motility (Wang et al., 2011a; Yu et al., 2011). Conversely, parkin mutations promote the degradation of DRP-1 through ubiquitylation (Wang et al., 2011b). In this same niche, mutations in MFN2 causes neurodegenerative diseases such as Charcot–Marie–Tooth disease (CMT) type 2A (CMT2A) (Cartoni and Martinou, 2009), which is characterized by axonal degeneration, muscular atrophy and peripheral neuropathy (Züchner et al., 2004). This study was further supported by a recent work showing that DRP-1 ubiquitylation and degradation was increased in CMT2A patients with the R364W MFN2 mutation, which promotes mitochondrial hyperfusion, suggesting that MFN2 regulates DRP-1 function (Das et al., 2021).

A good example of pathologies arising from dysfunctions in the Ca^{2+} signaling niche are mutations in Sig-1R, which can lead to neurodegenerative diseases such as ALS. Owing to its interactions with a variety of voltage and ion channels on this MAN, Sig-1R plays an important role in neuronal function and in promoting proteostasis. Indeed, loss-of-functions in Sig-1R impair its binding to IP_3R, resulting in IP_3R mislocalization, Ca^{2+} signaling disruption, a reduction in ER–mitochondria contacts and, importantly, accumulation of mutant superoxide dismutase 1, a key driver of familial forms of ALS, at ERMCSs (Al-Saif et al., 2011; Watanabe et al., 2016).

Alterations in the lipid exchange niche play an important role in ER stress induction in non-alcoholic steatohepatitis (NASH)-like steatosis by dynamically controlling phosphatidylserine transfer from the ER to mitochondria in hepatocytes (Sood et al., 2014). Interestingly, MFN2 has been shown to be downregulated in NASH models; however, overexpression of MFN2 is sufficient to reverse the effects of NASH, which might be due to MFN2 ability to bind PS and transfer it from the ER into the mitochondria, thereby providing protection against NASH-like phenotypes and liver cancer (Hernández-Alvarez et al., 2019).

Metabolic diseases can also arise from dysfunctions in several MANs. For example, the insulin secretion pathway depends on proper lipid transport from the ER to the mitochondria by enzymes such as diacylglycerol acetyltransferase 2, which converts diacylglycerols into triglycerides. Impaired lipid conversion at the lipid exchange niche leads to the accumulation of diacylglycerols and thus repression of the insulin receptor (Lowell and Shulman, 2005). In addition, disruption of other MAN-associated processes, such as Ca^{2+} exchange and UPR, have also been associated with insulin resistance (Cheng et al., 2020).

Mitochondrial–lysosomal niches have also been implicated in neurodegenerative diseases, such as PD, resulting from PINK1 and Parkin mutations, which result in mitochondrial defects and neuronal loss (Park et al., 2018; Yambire et al., 2020). Metabolic disorders, such as the lysosomal storage disease (LSD) Niemann–Pick type C (NPC), might also result from defects in Ca^{2+} signaling and lipid accumulation in mitochondrial–lysosomal niches (Annunziata et al., 2018). Finally, the neurodegenerative LSD GM1 gangliosidosis has been shown to lead to neuronal degeneration through ER Ca^{2+} depletion and activation of the UPR (Caciotti et al., 2011; Jeyakumar et al., 2003; Ledeen and Wu, 2006). Overall, these studies further underscore the role of various MANs in maintaining health and inducing disease through multiple alterations in the communication between the mitochondria and other organelles, such as the ER and lysosomes.

Concluding remarks

Recent advances in our understanding of cellular and organelle biology have allowed us to better understand the complexity of the interactions between different intracellular compartments and to consider the importance of studying the biology of the cell as a highly interconnected system. The development of new tools such as proximity-based fluorescent probes and novel super-resolution

microscopes have made it possible to unveil the intricacies of the signaling pathways involved in the physical and functional interaction between mitochondria and other organelles, such as the ER and lysosomes. Likewise, these tools might open the door to the development of novel therapeutic strategies that in the future could help in the treatment of cancer, metabolic pathologies and neurological diseases by targeting dysfunctions of interorganelle interfaces (Carreras-Sureda et al., 2017; Christ et al., 2020; Saito and Imaizumi, 2018). We regard each of these highly regulated interfaces between organelles as a niche, a signaling hub with functional relevance for organelle and cell physiology. In this Review, we have attempted to promote the new concept of MANs as a means to reference our understanding of the cellular mechanisms that regulate the functional and structural interactions between mitochondria and other organelles in health and disease. For example, important integrative cellular responses such as the UPR, mitochondrial membrane dynamics, Ca^{2+} signaling, lipid exchange and autophagy are regulated by MANs. In this line, proteins identified biochemically as belonging to a MAM are not necessarily part of a functional signaling pathway and do not necessarily configure a 'signaling niche'. For example, REEP1, a protein that regulates ER tubular morphology has been found in ERMCSs (Lim et al., 2015; Wang et al., 2021), but so far this localization of REEP1 is not associated with a signaling pathway; it is simply found at these contact sites because of superimposition of different organelles membranes. In contrast, bona fide MAN proteins are part of a functional mechanism that occurs through the association of two or more organelles and associated proteins, lipids and metabolites. Naturally, there is an overlap between the factors that can be categorized under both concepts, and, as new signaling pathways are uncovered, more molecules could be reassigned from being only physically located at a mitochondrial membrane to being part of a functional niche. Therefore, the concept of MANs proposed here emphasizes the functional pathways that emerge from exchanges between two or more organelles, as we believe that proteins belonging to these niches are prime candidates for studies aimed at understanding how these signaling pathways are related to cell physiology, and thus health and disease. Although knowledge in this field has increased in recent years, an integrative understanding that could allow the specific modulation of molecules present in MANs with the aim to determine their impact on the severity of various pathologies is still missing. Therefore, by evolving our perspective of intracellular communication, we are also hopefully improving the landscape for drug discovery and therapeutic interventions.

Competing interests
The authors declare no competing or financial interests.

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References
Adachi, Y., Itoh, K., Yamada, T., Ramachandran, R., Iijima, M., Correspondence, H. S., Cerveny, K. L., Suzuki, T. L., Macdonald, P., Frohman, M. A. et al. (2016). Coincident phosphatidic acid interaction restrains Drp1 in mitochondrial division. *Mol. Cell* **63**, 1034-1043. doi:10.1016/j.molcel.2016.08.013

Akl, H., Monaco, G., La Rovere, R., Welkenhuyzen, K., Kiviluoto, S., Vervliet, T., Molgó, J., Distelhorst, C. W., Missiaen, L., Mikoshiba, K. et al. (2013). IP3R2 levels dictate the apoptotic sensitivity of diffuse large B-cell lymphoma cells to an IP3R-derived peptide targeting the BH4 domain of Bcl-2. *Cell Death Dis.* **4**, e362. doi:10.1038/cddis.2013.140

Alpy, F., Rousseau, A., Schwab, Y., Legueux, F., Stoll, I., Wendling, C., Spiegelhalter, C., Kessler, P., Mathelin, C., Rio, M.-C. et al. (2013). STARD3 or STARD3NL and VAP form a novel molecular tether between late endosomes and the ER. *J. Cell Sci.* **126**, 5500-5512. doi:10.1242/jcs.139295

Al-Saif, A., Al-Mohanna, F. and Bohlega, S. (2011). A mutation in sigma-1 receptor causes juvenile amyotrophic lateral sclerosis. *Ann. Neurol.* **70**, 913-919. doi:10.1002/ana.22534

Annunziata, I., Sano, R. and D'Azzo, A. (2018). Mitochondria-associated ER membranes (MAMs) and lysosomal storage diseases. *Cell Death Dis.* **9**, 328. doi:10.1038/s41419-017-0025-4

Arasaki, K., Shimizu, H., Mogari, H., Nishida, N., Hirota, N., Furuno, A., Kudo, Y., Baba, M., Baba, N., Cheng, J. et al. (2015). A role for the ancient SNARE syntaxin 17 in regulating mitochondrial division. *Dev. Cell* **32**, 304-317. doi:10.1016/j.devcel.2014.12.011

Balsa, E., Soustek, M. S., Thomas, A., Cogliati, S., García-Poyatos, C., Martín-García, E., Jedrychowski, M., Gygi, S. P., Enriquez, J. A. and Puigserver, P. (2019). ER and nutrient stress promote assembly of respiratory chain supercomplexes through the PERK-eIF2α axis. *Mol. Cell* **74**, 877-890.e6. doi:10.1016/j.molcel.2019.03.031

Baines, C. P., Kaiser, R. A., Purcell, N. H., Blair, N. S., Osinska, H., Hambleton, M. A., Brunskill, E. W., Sayen, M. R., Gottlieb, R. A., Dorn, G. W., et al. (2005). Loss of cyclophilin D reveals a critical role for mitochondrial permeability transition in cell death. *Nature* **434**, 658-662. doi:10.1038/nature03434

Biazik, J., Ylä-Anttila, P., Vihinen, H., Jokitalo, E. and Eskelinen, E.-L. (2015). Ultrastructural relationship of the phagophore with surrounding organelles. *Autophagy* **11**, 439-451. doi:10.1080/15548627.2015.1017178

Bittremieux, M., La Rovere, R. M., Akl, H., Martines, C., Welkenhuyzen, K., Dubron, K., Baes, M., Janssens, A., Vandenberghe, P., Laurenti, L. et al. (2019). Constitutive IP3 signaling underlies the sensitivity of B-cell cancers to the Bcl-2/IP3 receptor disruptor BIRD-2. *Cell Death Differ.* **26**, 531-547. doi:10.1038/s41418-018-0142-3

Booth, D. M., Várnai, P., Joseph, S. K. and Hajnóczky, G. (2021). Oxidative bursts of single mitochondria mediate retrograde signaling toward the ER. *Mol. Cell* **81**, 3866-3876.e2. doi:10.1016/j.molcel.2021.07.014

Caciotti, A., Garman, S. C., Rivera-Colón, Y., Procopio, E., Catarzi, S., Ferri, L., Guido, C., Martelli, P., Parini, R., Antuzzi, D. et al. (2011). GM1 gangliosidosis and Morquio B disease: an update on genetic alterations and clinical findings. *Biochim. Biophys. Acta* **1812**, 782-790. doi:10.1016/j.bbadis.2011.03.018

Calfon, M., Zeng, H., Urano, F., Till, J. H., Hubbard, S. R., Harding, H. P., Clark, S. G. and Ron, D. (2002). IRE1 couples endoplasmic reticulum load to secretory capacity by processing the XBP-1 mRNA. *Nature* **415**, 92-96. doi:10.1038/415092a

Cárdenas, C., Miller, R. A., Smith, I., Bui, T., Molgó, J., Müller, M., Vais, H., Cheung, K.-H., Yang, J., Parker, I. et al. (2010). Essential regulation of cell bioenergetics by constitutive InsP3 receptor Ca2+ transfer to mitochondria. *Cell* **142**, 270-283. doi:10.1016/j.cell.2010.06.007

Carpio, M. A., Means, R. E., Brill, A. L., Sainz, A., Ehrlich, B. E. and Katz, S. G. (2021). BOK controls apoptosis by Ca2+ transfer through ER-mitochondrial contact sites. *Cell Rep.* **34**, 108827. doi:10.1016/j.celrep.2021.108827

Carreras-Sureda, A., Pihán, P. and Hetz, C. (2017). The unfolded protein response: at the intersection between endoplasmic reticulum function and mitochondrial bioenergetics. *Front. Oncol.* **7**, 55. doi:10.3389/fonc.2017.00055

Carreras-Sureda, A., Jaña, F., Urra, H., Durand, S., Mortenson, D. E., Sagredo, A., Bustos, G., Hazari, Y., Ramos-Fernández, E., Sassano, M. L. et al. (2019). Non-canonical function of IRE1α determines mitochondria-associated endoplasmic reticulum composition to control calcium transfer and bioenergetics. *Nat. Cell Biol.* **21**, 755-767. doi:10.1038/s41556-019-0329-y

Cartoni, R. and Martinou, J.-C. (2009). Role of mitofusin 2 mutations in the physiopathology of Charcot–Marie–Tooth disease type 2A. *Exp. Neurol.* **218**, 268-273. doi:10.1016/j.expneurol.2009.05.003

Cerqua, C., Anesti, V., Pyakurel, A., Liu, D., Naon, D., Wiche, G., Baffa, R., Dimmer, K. S. and Scorrano, L. (2010). Trichoplein/mitostatin regulates endoplasmic reticulum–mitochondria juxtaposition. *EMBO Rep.* **11**, 854-860. doi:10.1038/embor.2010.151

Chan, D. C. (2020). Mitochondrial dynamics and its involvement in disease. *Annu. Rev. Pathol. Mech. Dis.* **15**, 235-259. doi:10.1146/annurev-pathmechdis-012419-032711

Chen, J., Ying, H., Liu, X., Gu, J., Feng, R., Chen, T., Gao, H. and Wu, J. (2021). A transfer learning based super-resolution microscopy for biopsy slice images: the joint methods perspective. *IEEE/ACM Trans. Comput. Biol. Bioinf.* **18**, 103-113. doi:10.1109/TCBB.2020.2991173

Cheng, H., Gang, X., He, G., Liu, Y., Wang, Y., Zhao, X. and Wang, G. (2020). The molecular mechanisms underlying mitochondria-associated endoplasmic reticulum membrane-induced insulin resistance. *Front. Endocrinol.* **11**, 592129. doi:10.3389/fendo.2020.592129

Chlystun, M., Campanella, M., Law, A.-L., Duchen, M. R., Fatimathas, L., Levine, T. P., Gerke, V. and Moss, S. E. (2013). Regulation of mitochondrial morphogenesis by annexin A6. *PLoS One* **8**, e53774. doi:10.1371/journal.pone.0053774

Christ, M. G., Clement, A. M. and Behl, C. (2020). The sigma-1 receptor at the crossroad of proteostasis, neurodegeneration, and autophagy. *Trends Neurosci.* **43**, 79-81. doi:10.1016/j.tins.2019.12.002

Claypool, S. M. and Koehler, C. M. (2012). The complexity of cardiolipin in health and disease. *Trends Biochem. Sci.* **37**, 32-41. doi:10.1016/j.tibs.2011.09.003

Credle, J. J., Finer-Moore, J. S., Papa, F. R., Stroud, R. M. and Walter, P. (2005). On the mechanism of sensing unfolded protein in the endoplasmic reticulum. *Proc. Natl. Acad. Sci. USA* **102**, 18773-18784. doi:10.1073/pnas.0509487102

Das, R., Kamal, I. M., Das, S., Chakrabarti, S. and Chakrabarti, O. (2021). MITOL-mediated DRP1 ubiquitylation and degradation promotes mitochondrial hyperfusion in CMT2A-linked MFN2 mutant. *J. Cell Sci.* **135**, jcs257808. doi:10.1242/jcs.257808

De La Fuente, S., Fonteriz, R. I., Montero, M. and Alvarez, J. (2013). Ca2+ homeostasis in the endoplasmic reticulum measured with a new low-Ca2+-affinity targeted aequorin. *Cell Calcium* **54**, 37-45. doi:10.1016/j.ceca.2013.04.001

De Vos, K. J., Mórotz, G. M., Stoica, R., Tudor, E. L., Lau, K.-F., Ackerley, S., Warley, A., Shaw, C. E. and Miller, C. C. J. (2012). VAPB interacts with the mitochondrial protein PTPIP51 to regulate calcium homeostasis. *Hum. Mol. Genet.* **21**, 1299-1311. doi:10.1093/hmg/ddr559

Del Dotto, V., Fogazza, M., Carelli, V., Rugolo, M. and Zanna, C. (2018). Eight human OPA1 isoforms, long and short: what are they for? *Biochim. Acta Bioenerg.* **1859**, 263-269. doi:10.1016/j.bbabio.2018.01.005

Dikic, I. and Elazar, Z. (2018). Mechanism and medical implications of mammalian autophagy. *Nat. Rev. Mol. Cell Biol.* **19**, 349-364. doi:10.1038/s41580-018-0003-4

Dong, Z., Shanmughapriya, S., Tomar, D., Siddiqui, N., Lynch, S., Nemani, N., Breves, S. L., Zhang, X., Tripathi, A., Palaniappan, P. et al. (2017). Mitochondrial Ca2+ uniporter is a mitochondrial luminal redox sensor that augments MCU channel activity. *Mol. Cell* **65**, 1014-1028.e7. doi:10.1016/j.molcel.2017.01.032

Dulloo, I., Atakpa-Adaji, P., Yeh, Y.-C., Levet, C., Muliyil, S., Lu, F., Taylor, C. W. and Freeman, M. (2022). iRhom pseudoproteases regulate ER stress-induced cell death through IP3 receptors and BCL-2. *Nat. Commun.* **13**, 1257. doi:10.1038/s41467-022-28930-4

Echeverry, N., Bachmann, D., Ke, F., Strasser, A., Simon, H. U. and Kaufmann, T. (2013). Intracellular localization of the BCL-2 family member BOK and functional implications. *Cell Death Differ.* **20**, 785-799. doi:10.1038/cdd.2013.10

Faizan, M. I. and Ahmad, T. (2021). Altered mitochondrial calcium handling and cell death by necroptosis: an emerging paradigm. *Mitochondrion* **57**, 47-62. doi:10.1016/j.mito.2020.12.004

Fessler, E., Eckl, E. M., Schmitt, S., Mancilla, I. A., Meyer-Bender, M. F., Hanf, M., Philippou-Massier, J., Krebs, S., Zischka, H. and Jae, L. T. (2020). A pathway coordinated by DELE1 relays mitochondrial stress to the cytosol. *Nature* **579**, 433-437. doi:10.1038/s41586-020-2076-4

Fonseca, T. B., Sánchez-Guerrero, Á., Milosevic, I. and Raimundo, N. (2019). Mitochondrial fission requires DRP1 but not dynamins. *Nature* **570**, E34-E42. doi:10.1038/s41586-019-1296-y

Frank, S., Gaume, B., Bergmann-Leitner, E. S., Leitner, W. W., Robert, E. G., Catez, F., Smith, C. L. and Youle, R. J. (2001). The role of dynamin-related protein 1, a mediator of mitochondrial fission, in Apoptosis. *Dev. Cell* **1**, 515-525. doi:10.1016/S1534-5807(01)00055-7

Friedman, J. R., Lackner, L. L., West, M., Dibenedetto, J. R., Nunnari, J. and Voeltz, G. K. (2011). ER tubules mark sites of mitochondrial division. *Science* **334**, 358-362. doi:10.1126/science.1207385

Fujimoto, M., Hayashi, T. and Su, T.-P. (2012). The role of cholesterol in the association of endoplasmic reticulum membranes with mitochondria. *Biochem. Biophys. Res. Commun.* **417**, 635-639. doi:10.1016/j.bbrc.2011.12.022

Gao, S. and Hu, J. (2021). Mitochondrial fusion: the machineries in and out. *Trends Cell Biol.* **31**, 62-74. doi:10.1016/j.tcb.2020.09.008

Garofalo, T., Matarrese, P., Manganelli, V., Marconi, M., Tinari, A., Gambardella, L., Faggioni, A., Misasi, R., Sorice, M. and Malorni, W. (2016). Evidence for the involvement of lipid rafts localized at the ER-mitochondria associated membranes in autophagosome formation. *Autophagy* **12**, 917-935. doi:10.1080/15548627.2016.1160971

Garrity, A. G., Wang, W., Collier, C. M., Levey, S. A., Gao, Q. and Xu, H. (2016). The endoplasmic reticulum, not the pH gradient, drives calcium refilling of lysosomes. *Elife* **5**, e15887. doi:10.7554/eLife.15887

Giorgi, C., Marchi, S. and Pinton, P. (2018). The machineries, regulation and cellular functions of mitochondrial calcium. *Nat. Rev. Mol. Cell Biol.* **19**, 713-730. doi:10.1038/s41580-018-0052-8

Gomez-Suaga, P., Paillusson, S., Stoica, R., Noble, W., Hanger, D. P. and Miller, C. C. J. (2017). The ER-mitochondria tethering complex VAPB-PTPIP51 regulates autophagy. *Curr. Biol.* **27**, 371-385. doi:10.1016/j.cub.2016.12.038

Gray, M. W., Burger, G. and Lang, B. F. (1999). Mitochondrial evolution. *Science* **283**, 1476-1481. doi:10.1126/science.283.5407.1476

Green, D. R. (2019). The coming decade of cell death research: five riddles. *Cell* **177**, 1094-1107. doi:10.1016/j.cell.2019.04.024

Guillén-Samander, A., Leonzino, M., Hanna, M. G., Tang, N., Shen, H. and De Camilli, P. (2021). VPS13D bridges the ER to mitochondria and peroxisomes via Miro. *J. Cell Biol.* **220**, e202010004. doi:10.1083/jcb.202010004

Guo, X., Chen, K. H., Guo, Y., Liao, H., Tang, J. and Xiao, R. P. (2007). Mitofusin 2 triggers vascular smooth muscle cell apoptosis via mitochondrial death pathway. *Circ. Res.* **101**, 1113-1122. doi:10.1161/CIRCRESAHA.107.157644

Guo, X., Aviles, G., Liu, Y., Tian, R., Unger, B. A., Lin, Y. H. T., Wiita, A. P., Xu, K., Correia, M. A. and Kampmann, M. (2020). Mitochondrial stress is relayed to the cytosol by an OMA1–DELE1–HRI pathway. *Nature* **579**, 427-432. doi:10.1038/s41586-020-2078-2

Hailey, D. W., Rambold, A. S., Satpute-Krishnan, P., Mitra, K., Sougrat, R., Kim, P. K. and Lippincott-Schwartz, J. (2010). Mitochondria supply membranes for autophagosome biogenesis during starvation. *Cell* **141**, 656-667. doi:10.1016/j.cell.2010.04.009

Hamasaki, M., Furuta, N., Matsuda, A., Nezu, A., Yamamoto, A., Fujita, N., Oomori, H., Noda, T., Haraguchi, T., Hiraoka, Y. et al. (2013). Autophagosomes form at ER-mitochondria contact sites. *Nature* **495**, 389-393. doi:10.1038/nature11910

Hayashi, T. and Fujimoto, M. (2010). Detergent-resistant microdomains determine the localization of sigma-1 receptors to the endoplasmic reticulum-mitochondria junction. *Mol. Pharmacol.* **77**, 517-528. doi:10.1124/mol.109.062539

Hayashi, T. and Su, T.-P. (2004). Sigma-1 receptors at galactosylceramide-enriched lipid microdomains regulate oligodendrocyte differentiation. *Proc. Natl. Acad. Sci. USA* **101**, 14949-14954. doi:10.1073/pnas.0402890101

Hayashi-Nishino, M., Fujita, N., Noda, T., Yamaguchi, A., Yoshimori, T. and Yamamoto, A. (2009). A subdomain of the endoplasmic reticulum forms a cradle for autophagosome formation. *Nat. Cell Biol.* **11**, 1433-1437. doi:10.1038/ncb1991

Hernández-Alvarez, M. I., Sebastián, D., Vives, S., Ivanova, S., Bartoccioni, P., Kakimoto, P., Plana, N., Veiga, S. R., Hernández, V., Vasconcelos, N. et al. (2019). Deficient endoplasmic reticulum-mitochondrial phosphatidylserine transfer causes liver disease. *Cell* **177**, 881-895.e17. doi:10.1016/j.cell.2019.04.010

Herrera-Cruz, M. S. and Simmen, T. (2017). Of yeast, mice and men: MAMs come in two flavors. *Biol. Direct* **12**, 3. doi:10.1186/s13062-017-0174-5

Hetz, C., Zhang, K. and Kaufman, R. J. (2020). Mechanisms, regulation and functions of the unfolded protein response. *Nat. Rev. Mol. Cell Biol.* **21**, 421-438. doi:10.1038/s41580-020-0250-z

Hoppins, S., Edlich, F., Cleland, M., Banerjee, S., Mccaffery, J. M., Youle, R. and Nunnari, J. (2011). The soluble form of Bax regulates mitochondrial fusion via MFN2 homotypic complexes. *Mol. Cell* **41**, 150-160. doi:10.1016/j.molcel.2010.11.030

Hung, V., Lam, S. S., Udeshi, N. D., Svinkina, T., Guzman, G., Mootha, V. K., Carr, S. A. and Ting, A. Y. (2017). Proteomic mapping of cytosol-facing outer mitochondrial and ER membranes in living human cells by proximity biotinylation. *Elife* **6**, e24463. doi:10.7554/eLife.24463

Hwang, M.-S., Schwall, C. T., Pazarentzos, E., Datler, C., Alder, N. N. and Grimm, S. (2014). Mitochondrial Ca(2+) influx targets cardiolipin to disintegrate respiratory chain complex II for cell death induction. *Cell Death Differ.* **21**, 1733-1745. doi:10.1038/cdd.2014.84

Itakura, E., Kishi-Itakura, C. and Mizushima, N. (2012). The hairpin-type tail-anchored SNARE syntaxin 17 targets to autophagosomes for fusion with endosomes/lysosomes. *Cell* **151**, 1256-1269. doi:10.1016/j.cell.2012.11.001

Jenner, A., Peña–Blanco, A., Salvador–Gallego, R., Ugarte–Uribe, B., Zollo, C., Ganief, T., Bierlmeier, J., Mund, M., Lee, J. E., Ries, J. et al. (2022). DRP1 interacts directly with BAX to induce its activation and apoptosis. *EMBO J.* **41**, e108587. doi:10.15252/embj.2021108587

Jeyakumar, M., Thomas, R., Elliot-Smith, E., Smith, D. A., Van Der Spoel, A. C., D'Azzo, A., Perry, V. H., Butters, T. D., Dwek, R. A. and Platt, F. M. (2003). Central nervous system inflammation is a hallmark of pathogenesis in mouse models of GM1 and GM2 gangliosidosis. *Brain* **126**, 974-987. doi:10.1093/brain/awg089

John Peter, A. T., Petrungaro, C., Peter, M. and Kornmann, B. (2022). METALIC reveals interorganelle lipid flux in live cells by enzymatic mass tagging. *Nat. Cell Biol.* **24**, 996-1004. doi:10.1038/s41556-022-00917-9

Kagan, V. E., Tyurin, V. A., Jiang, J., Tyurina, Y. Y., Ritov, V. B., Amoscato, A. A., Osipov, A. N., Belikova, N. A., Kapralov, A. A., Kini, V. et al. (2005). Cytochrome C acts as A cardiolipin oxygenase required for release of proapoptotic factors. *Nat. Chem. Biol.* **1**, 223-232. doi:10.1038/nchembio727

Kale, J., Osterlund, E. J. and Andrews, D. W. (2018). BCL-2 family proteins: changing partners in the dance towards death. *Cell Death Differ.* **25**, 65-80. doi:10.1038/cdd.2017.186

Kameoka, S., Adachi, Y., Okamoto, K., Iijima, M. and Sesaki, H. (2018). Phosphatidic acid and cardiolipin coordinate mitochondrial dynamics. *Trends Cell Biol.* **28**, 67-76. doi:10.1016/j.tcb.2017.08.011

Kamerkar, S. C., Kraus, F., Sharpe, A. J., Pucadyil, T. J. and Ryan, M. T. (2018). Dynamin-related protein 1 has membrane constricting and severing abilities sufficient for mitochondrial and peroxisomal fission. *Nat. Commun.* **9**, 5239. doi:10.1038/s41467-018-07543-w

Karbowski, M., Lee, Y.-J., Gaume, B., Jeong, S.-Y., Frank, S., Nechushtan, A., Santel, A., Fuller, M., Smith, C. L. and Youle, R. J. (2002). Spatial and temporal association of Bax with mitochondrial fission sites, Drp1, and Mfn2 during apoptosis. *J. Cell Biol.* **159**, 931-938. doi:10.1083/jcb.200209124

Kaufman, R. J. (2002). Orchestrating the unfolded protein response in health and disease. *J. Clin. Investig.* **110**, 1389-1398. doi:10.1172/JCI0216886

Kim, H.-K., Lee, G.-H., Bhattarai, K. R., Lee, M.-S., Back, S. H., Kim, H.-R. and Chae, H.-J. (2021a). TMBIM6 (transmembrane BAX inhibitor motif containing 6) enhances autophagy through regulation of lysosomal calcium. *Autophagy* **17**, 761-778. doi:10.1080/15548627.2020.1732161

Kim, S., Wong, Y. C., Gao, F. and Krainc, D. (2021b). Dysregulation of mitochondria-lysosome contacts by GBA1 dysfunction in dopaminergic neuronal models of Parkinson's disease. *Nat. Commun.* **12**, 1807. doi:10.1038/s41467-021-22113-3

Kim, H., Lee, S., Jun, Y. and Lee, C. (2022). Structural basis for mitoguardin-2 mediated lipid transport at ER-mitochondrial membrane contact sites. *Nat. Commun.* **13**, 3702. doi:10.1038/s41467-022-31462-6

Kleele, T., Rey, T., Winter, J., Zaganelli, S., Mahecic, D., Perreten Lambert, H., Ruberto, F. P., Nemir, M., Wai, T., Pedrazzini, T. et al. (2021). Distinct fission signatures predict mitochondrial degradation or biogenesis. *Nature* **593**, 435-439. doi:10.1038/s41586-021-03510-6

Kornmann, B., Currie, E., Collins, S. R., Schuldiner, M., Nunnari, J., Weissman, J. S. and Walter, P. (2009). An ER-mitochondria tethering complex revealed by a synthetic biology screen. *Science* **325**, 477-481. doi:10.1126/science.1175088

Korobova, F., Ramabhadran, V. and Higgs, H. N. (2013). An actin-dependent step in mitochondrial fission mediated by the ER-associated formin INF2. *Science* **339**, 1228360. doi:10.1126/science.1228360

Korobova, F., Gauvin, T. J. and Higgs, H. N. (2014). A role for myosin II in mammalian mitochondrial fission. *Curr. Biol.* **24**, 409-414. doi:10.1016/j.cub.2013.12.032

Kozlov, G. and Gehring, K. (2020). Calnexin cycle – structural features of the ER chaperone system. *FEBS J.* **287**, 4322-4340. doi:10.1111/febs.15330

Latorre-Muro, P., O'Malley, K. E., Bennett, C. F., Perry, E. A., Balsa, E., Tavares, C. D. J., Jedrychowski, M., Gygi, S. P. and Puigserver, P. (2021). A cold-stress-inducible PERK/OGT axis controls TOM70-assisted mitochondrial protein import and cristae formation. *Cell Metab.* **33**, 598-614.e7. doi:10.1016/j.cmet.2021.01.013

Lazcano, A. and Peretó, J. (2017). On the origin of mitosing cells: a historical appraisal of Lynn Margulis endosymbiotic theory. *J. Theor. Biol.* **434**, 80-87. doi:10.1016/j.jtbi.2017.06.036

Lebeau, J., Saunders, J. M., Moraes, V. W. R., Madhavan, A., Madrazo, N., Anthony, M. C. and Wiseman, R. L. (2018). The PERK arm of the unfolded protein response regulates mitochondrial morphology during acute endoplasmic reticulum stress. *Cell Reports* **22**, 2827-2836. doi:10.1016/j.celrep.2018.02.055

Ledeen, R. W. and Wu, G. (2006). GM1 ganglioside: another nuclear lipid that modulates nuclear calcium. GM1 potentiates the nuclear sodium-calcium exchanger. *Can. J. Physiol. Pharmacol.* **84**, 393-402. doi:10.1139/y05-133

Lee, J. E., Westrate, L. M., Wu, H., Page, C. and Voeltz, G. (2016). Multiple dynamin family members collaborate to drive mitochondrial division. *Mol. Biol. Cell.* **26**, 139-143. doi:10.1038/nature20555

Levy, J. M. M., Towers, C. G. and Thorburn, A. (2017). Targeting autophagy in cancer. *Nat. Rev. Cancer* **17**, 528-542. doi:10.1038/nrc.2017.53

Lewis, S. C., Uchiyama, L. F. and Nunnari, J. (2016). ER-mitochondria contacts couple mtDNA synthesis with mitochondrial division in human cells. *Science* **353**, 261-269. doi:10.1126/science.aaf5549

Li, C., Wang, X., Vais, H., Thompson, C. B., Foskett, J. K. and White, C. (2007). Apoptosis regulation by Bcl-x(L) modulation of mammalian inositol 1,4,5-trisphosphate receptor channel isoform gating. *Proc. Natl. Acad. Sci. USA* **104**, 12565-12570. doi:10.1073/pnas.0702489104

Lim, Y., Cho, I.-T., Schoel, L. J., Cho, G. and Golden, J. A. (2015). Hereditary spastic paraplegia-linked REEP1 modulates endoplasmic reticulum/mitochondria contacts. *Ann. Neurol.* **78**, 679-696. doi:10.1002/ana.24488

Llambi, F., Wang, Y.-M., Victor, B., Yang, M., Schneider, D. M., Gingras, S., Parsons, M. J., Zheng, J. H., Brown, S. A., Pelletier, S. et al. (2016). BOK is a non-canonical BCL-2 family effector of apoptosis regulated by ER-associated degradation. *Cell* **165**, 421-433. doi:10.1016/j.cell.2016.02.026

Lowell, B. B. and Shulman, G. I. (2005). Mitochondrial dysfunction and type 2 diabetes. *Science* **307**, 384-387. doi:10.1126/science.1104343

Macdonald, P. J., Stepanyants, N., Mehrotra, N., Mears, J. A., Qi, X., Sesaki, H. and Ramachandran, R. (2014). A dimeric equilibrium intermediate nucleates Drp1 reassembly on mitochondrial membranes for fission. *Mol. Biol. Cell* **25**, 1905-1915. doi:10.1091/mbc.e14-02-0728

Manor, U., Bartholomew, S., Golani, G., Christenson, E., Kozlov, M., Higgs, H., Spudich, J. and Lippincott-Schwartz, J. (2015). A mitochondria-anchored isoform of the actin-nucleating spire protein regulates mitochondrial division. *Elife* **4**, e08828. doi:10.7554/eLife.08828

Marchi, S. and Pinton, P. (2014). The mitochondrial calcium uniporter complex: molecular components, structure and physiopathological implications. *J. Physiol.* **592**, 829-839. doi:10.1113/jphysiol.2013.268235

Marchi, S., Patergnani, S., Missiroli, S., Morciano, G., Rimessi, A., Wieckowski, M. R., Giorgi, C. and Pinton, P. (2018). Mitochondrial and endoplasmic reticulum calcium homeostasis and cell death. *Cell Calcium* **69**, 62-72. doi:10.1016/j.ceca.2017.05.003

Mattenberger, Y., James, D. I. and Martinou, J. C. (2003). Fusion of mitochondria in mammalian cells is dependent on the mitochondrial inner membrane potential and independent of microtubules or actin. *FEBS Lett.* **538**, 53-59. doi:10.1016/S0014-5793(03)00124-8

Meneses-Salas, E., García-Melero, A., Kanerva, K., Blanco-Muñoz, P., Morales-Paytuvi, F., Bonjoch, J., Casas, J., Egert, A., Beevi, S. S., Jose, J. et al. (2020). Annexin A6 modulates TBC1D15/Rab7/StARD3 axis to control endosomal cholesterol export in NPC1 cells. *Cell. Mol. Life Sci.* **77**, 2839-2857. doi:10.1007/s00018-019-03330-y

Milani, M., Byrne, D. P., Greaves, G., Butterworth, M., Cohen, G. M., Eyers, P. A. and Varadarajan, S. (2017). DRP-1 is required for BH3 mimetic-mediated mitochondrial fragmentation and apoptosis. *Cell Death Dis.* **8**, e2552. doi:10.1038/cddis.2016.485

Milani, M., Beckett, A. J., Al-Zebeeby, A., Luo, X., Prior, I. A., Cohen, G. M. and Varadarajan, S. (2019). DRP-1 functions independently of mitochondrial structural perturbations to facilitate BH3 mimetic-mediated apoptosis. *Cell Death Discov.* **5**, 117. doi:10.1038/s41420-019-0199-x

Montessuit, S., Somasekharan, S. P., Terrones, O., Lucken-Ardjomande, S., Herzig, S., Schwarzenbacher, R., Manstein, D. J., Bossy-Wetzel, E., Basañez, G., Meda, P. et al. (2010). Membrane remodeling induced by the dynamin-related protein Drp1 stimulates bax oligomerization. *Cell* **142**, 889-901. doi:10.1016/j.cell.2010.08.017

Mori, T., Hayashi, T., Hayashi, E. and Su, T. P. (2013). Sigma-1 receptor chaperone at the ER-mitochondrion interface mediates the mitochondrion-ER-nucleus signaling for cellular survival. *PLoS One* **8**, e76941. doi:10.1371/journal.pone.0076941

Muñoz, J. P., Ivanova, S., Sánchez-Wandelmer, J., Martínez-Cristóbal, P., Noguera, E., Sancho, A., Díaz-Ramos, A., Hernández-Alvarez, M. I., Sebastián, D., Mauvezin, C. et al. (2013). Mfn2 modulates the UPR and mitochondrial function via repression of PERK. *EMBO J.* **32**, 2348-2361. doi:10.1038/emboj.2013.168

Myhill, N., Lynes, E. M., Nanji, J. A., Blagoveshchenskaya, A. D., Fei, H., Simmen, K. C., Cooper, T. J., Thomas, G. and Simmen, T. (2008). The subcellular distribution of calnexin is mediated by PACS-2. *Mol. Biol. Cell* **19**, 2777-2788. doi:10.1091/mbc.e07-10-0995

Nagashima, S., Tábara, L.-C., Tilokani, L., Paupe, V., Anand, H., Pogson, J. H., Zunino, R., Mcbride, H. M. and Prudent, J. (2020). Golgi-derived PI(4)P-containing vesicles drive late steps of mitochondrial division. *Science* **367**, 1366-1371. doi:10.1126/science.aax6089

Naon, D., Zaninello, M., Giacomello, M., Varanita, T., Grespi, F., Lakshminaranayan, S., Serafini, A., Semenzato, M., Herkenne, S., Hernández-Alvarez, M. I. et al. (2016). Critical reappraisal confirms that Mitofusin 2 is an endoplasmic reticulum–mitochondria tether. *Proc. Natl. Acad. Sci. USA* **113**, 11249-11254. doi:10.1073/pnas.1606786113

Nemani, N., Carvalho, E., Tomar, D., Dong, Z., Ketschek, A., Breves, S. L., Jaña, F., Worth, A. M., Heffler, J., Palaniappan, P. et al. (2018). MIRO-1 determines mitochondrial shape transition upon GPCR activation and Ca2+ stress. *Cell Rep.* **23**, 1005-1019. doi:10.1016/j.celrep.2018.03.098

Ngoh, G. A., Papanicolaou, K. N. and Walsh, K. (2012). Loss of mitofusin 2 promotes endoplasmic reticulum stress. *J. Biol. Chem.* **287**, 20321-20332. doi:10.1074/jbc.M112.359174

Nishimura, T. and Tooze, S. A. (2020). Emerging roles of ATG proteins and membrane lipids in autophagosome formation. *Cell Discov.* **6**, 32. doi:10.1038/s41421-020-0161-3

Nishimura, T., Tamura, N., Kono, N., Shimanaka, Y., Arai, H., Yamamoto, H. and Mizushima, N. (2017). Autophagosome formation is initiated at phosphatidylinositol synthase–enriched ER subdomains. *EMBO J.* **36**, 1719-1735. doi:10.15252/embj.201695189

Onoue, K., Jofuku, A., Ban-Ishihara, R., Ishihara, T., Maeda, M., Koshiba, T., Itoh, T., Fukuda, M., Otera, H., Oka, T. et al. (2013). Fis1 acts as a mitochondrial recruitment factor for TBC1D15 that is involved in regulation of mitochondrial morphology. *J. Cell Sci.* **126**, 176-185. doi:10.1242/jcs.111211

Otera, H., Miyata, N., Kuge, O. and Mihara, K. (2016). Drp1-dependent mitochondrial fission via MiD49/51 is essential for apoptotic cristae remodeling. *J. Cell Biol.* **212**, 531-544. doi:10.1083/jcb.201508099

Palmer, C. S., Elgass, K. D., Parton, R. G., Osellame, L. D., Stojanovski, D. and Ryan, M. T. (2013). Adaptor proteins MiD49 and MiD51 can act independently of Mff and Fis1 in Drp1 recruitment and are specific for mitochondrial fission. *J. Biol. Chem.* **288**, 27584-27593. doi:10.1074/jbc.M113.479873

Park, J.-S., Davis, R. L. and Sue, C. M. (2018). Mitochondrial dysfunction in Parkinson's disease: new mechanistic insights and therapeutic perspectives. *Curr. Neurol. Neurosci. Rep.* **18**, 21. doi:10.1007/s11910-018-0829-3

Peng, W., Wong, Y. C. and Krainc, D. (2020). Mitochondria-lysosome contacts regulate mitochondrial Ca^{2+} dynamics via lysosomal TRPML1. *Proc. Natl. Acad. Sci. USA* **117**, 19266-19275. doi:10.1073/pnas.2003236117

Pihán, P., Carreras-Sureda, A. and Hetz, C. (2017). BCL-2 family: integrating stress responses at the ER to control cell demise. *Cell Death Differ.* **24**, 1478-1487. doi:10.1038/cdd.2017.82

Pihán, P., Lisbona, F., Borgonovo, J., Edwards-Jorquera, S., Nunes-Hasler, P., Castillo, K., Kepp, O., Urra, H., Saarnio, S., Vihinen, H., et al. (2021). Control of

lysosomal-mediated cell death by the pH-dependent calcium channel RECS1. *Sci. Adv.* **7**, eabe5469. doi:10.1126/sciadv.abe5469
Pihán, P., Milani, M. and Hetz, C. (2022). Proapoptotic RECS1: a requisite gateway to lysosomal dysfunction and death. *Neural Regen. Res.* **17**, 2695-2696. doi:10.4103/1673-5374.339487
Pinton, P., Ferrari, D., Rapizzi, E., Di Virgilio, F., Pozzan, T. and Rizzuto, R. (2001). The Ca2+ concentration of the endoplasmic reticulum is a key determinant of ceramide-induced apoptosis: significance for the molecular mechanism of Bcl-2 action. *EMBO J.* **20**, 2690-2701. doi:10.1093/emboj/20.11.2690
Prinz, W. A., Toulmay, A. and Balla, T. (2020). The functional universe of membrane contact sites. *Nat. Rev. Mol. Cell Biol.* **21**, 7-24. doi:10.1038/s41580-019-0180-9
Prudent, J. and Mcbride, H. M. (2017). The mitochondria-endoplasmic reticulum contact sites: a signalling platform for cell death. *Curr. Opin. Cell Biol.* **47**, 52-63. doi:10.1016/j.ceb.2017.03.007
Prudent, J., Zunino, R., Sugiura, A., Mattie, S., Shore, G. C. and Mcbride, H. M. (2015). MAPL SUMOylation of Drp1 stabilizes an ER/mitochondrial platform required for cell death. *Mol. Cell* **59**, 941-955. doi:10.1016/j.molcel.2015.08.001
Rizzuto, R., Pinton, P., Carrington, W., Fay, F. S., Fogarty, K. E., Lifshitz, L. M., Tuft, R. A. and Pozzan, T. (1998). Close contacts with the endoplasmic reticulum as determinants of mitochondrial Ca2+ responses. *Science* **280**, 1763-1766. doi:10.1126/science.280.5370.1763
Rojas-Rivera, D. and Hetz, C. (2015). TMBIM protein family: ancestral regulators of cell death. *Oncogene* **34**, 269-280. doi:10.1038/onc.2014.6
Ron, D. and Walter, P. (2007). Signal integration in the endoplasmic reticulum unfolded protein response. *Nat. Rev. Mol. Cell Biol.* **8**, 519-529. doi:10.1038/nrm2199
Rosa, N., Ivanova, H., Wagner, L. E., Kale, J., La Rovere, R., Welkenhuyzen, K., Louros, N., Karamanou, S., Shabardina, V., Lemmens, I. et al. (2022). Bcl-xL acts as an inhibitor of IP3R channels, thereby antagonizing Ca2+-driven apoptosis. *Cell Death Differ.* **29**, 788-805. doi:10.1038/s41418-021-00894-w
Saito, A. and Imaizumi, K. (2018). Unfolded protein response-dependent communication and contact among endoplasmic reticulum, mitochondria, and plasma membrane. *Int. J. Mol. Sci.* **19**, 3215. doi:10.3390/ijms19103215
Sano, R., Annunziata, I., Patterson, A., Moshiach, S., Gomero, E., Opferman, J., Forte, M. and D'Azzo, A. (2009). GM1-ganglioside accumulation at the mitochondria-associated ER membranes links ER stress to Ca2+-dependent mitochondrial apoptosis. *Mol. Cell* **36**, 500-511. doi:10.1016/j.molcel.2009.10.021
Schrader, M., Costello, J. L., Godinho, L. F., Azadi, A. S. and Islinger, M. (2016). Proliferation and fission of peroxisomes — an update. *Biochim. Biophys. Acta Mol. Cell Res.* **1863**, 971-983. doi:10.1016/j.bbamcr.2015.09.024
Schulman, J. J., Wright, F. A., Kaufmann, T. and Wojcikiewicz, R. J. H. (2013). The Bcl-2 protein family member Bok binds to the coupling domain of inositol 1,4,5-trisphosphate receptors and protects them from proteolytic cleavage. *J. Biol. Chem.* **288**, 25340-25349. doi:10.1074/jbc.M113.496570
Scorrano, L., De Matteis, M. A., Emr, S., Giordano, F., Hajnóczky, G., Kornmann, B., Lackner, L. L., Levine, T. P., Pellegrini, L., Reinisch, K. et al. (2019). Coming together to define membrane contact sites. *Nat. Commun.* **10**, 1287. doi:10.1038/s41467-019-09253-3
Sebastián, D., Hernández-Alvarez, M. I., Segalés, J., Sorianello, E., Muñoz, J. P., Sala, D., Waget, A., Liesa, M., Paz, J. C., Gopalacharyulu, P. et al. (2012). Mitofusin 2 (Mfn2) links mitochondrial and endoplasmic reticulum function with insulin signaling and is essential for normal glucose homeostasis. *Proc. Natl. Acad. Sci. U.S.A.* **109**, 5523-5528. doi:10.1073/pnas.1108220109
Sigal, Y. M., Zhou, R. and Zhuang, X. (2018). Visualizing and discovering cellular structures with super-resolution microscopy. *Science* **361**, 880-887. doi:10.1126/science.aau1044
Simmen, T., Aslan, J. E., Blagoveshchenskaya, A. D., Thomas, L., Wan, L., Xiang, Y., Feliciangeli, S. F., Hung, C.-H., Crump, C. M. and Thomas, G. (2005). PACS-2 controls endoplasmic reticulum–mitochondria communication and Bid-mediated apoptosis. *EMBO J.* **24**, 717. doi:10.1038/sj.emboj.7600559
Singh, R., Letai, A. and Sarosiek, K. (2019). Regulation of apoptosis in health and disease: the balancing act of BCL-2 family proteins. *Nat. Rev. Mol. Cell Biol.* **20**, 175-193. doi:10.1038/s41580-018-0089-8
Somlyo, A. P. (1984). Cellular site of calcium regulation. *Nature* **309**, 516-517. doi:10.1038/309516b0
Sood, A., Jeyaraju, D. V., Prudent, J., Caron, A., Lemieux, P., Mcbride, H. M., Laplante, M., Tóth, K. and Pellegrini, L. (2014). A Mitofusin-2-dependent inactivating cleavage of Opa1 links changes in mitochondria cristae and ER contacts in the postprandial liver. *Proc. Natl. Acad. Sci. U.S.A.* **111**, 16017-16022. doi:10.1073/pnas.1408061111
Streb, H., Irvine, R. F., Berridge, M. J. and Schulz, I. (1983). Release of Ca2+ from a nonmitochondrial intracellular store in pancreatic acinar cells by inositol-1,4,5-trisphosphate. *Nature* **306**, 67-69. doi:10.1038/306067a0
Tábara, L.-C., Morris, J. L. and Prudent, J. (2021). The complex dance of organelles during mitochondrial division. *Trends Cell Biol.* **31**, 241-253. doi:10.1016/j.tcb.2020.12.005
Tang, Z., Takahashi, Y., He, H., Hattori, T., Chen, C., Liang, X., Chen, H., Young, M. M. and Wang, H.-G. (2019). TOM40 targets Atg2 to mitochondria-associated ER membranes for phagophore expansion. *Cell Rep.* **28**, 1744-1757.e5. doi:10.1016/j.celrep.2019.07.036
Van Der Bliek, A. M., Shen, Q. and Kawajiri, S. (2013). Mechanisms of mitochondrial fission and fusion. *Cold Spring Harbor Perspect. Biol.* **5**, a011072. doi:10.1101/cshperspect.a011072
Vance, J. E. (1990). Phospholipid synthesis in a membrane fraction associated with mitochondria. *J. Biol. Chem.* **265**, 7248-7256. doi:10.1016/S0021-9258(19)39106-9
Vance, J. E. (2008). Thematic review series: glycerolipids. phosphatidylserine and phosphatidylethanolamine in mammalian cells: two metabolically related aminophospholipids. *J. Lipid Res.* **49**, 1377-1387. doi:10.1194/jlr.R700020-JLR200
Vance, J. E. (2014). MAM (mitochondria-associated membranes) in mammalian cells: lipids and beyond. *Biochim. Biophys. Acta Mol. Cell Biol. Lipids* **1841**, 595-609. doi:10.1016/j.bbalip.2013.11.014
Verfaillie, T., Rubio, N., Garg, A. D., Bultynck, G., Rizzuto, R., Decuypere, J. P., Piette, J., Linehan, C., Gupta, S., Samali, A. et al. (2012). PERK is required at the ER-mitochondrial contact sites to convey apoptosis after ROS-based ER stress. *Cell Death Differ.* **19**, 1880-1891. doi:10.1038/cdd.2012.74
Voelker, D. R. (1989). Phosphatidylserine translocation to the mitochondrion is an ATP-dependent process in permeabilized animal cells. *Proc. Natl. Acad. Sci. USA* **86**, 9921-9925. doi:10.1073/pnas.86.24.9921
Wang, X., Winter, D., Ashrafi, G., Schlehe, J., Wong, Y. L., Selkoe, D., Rice, S., Steen, J., Lavoie, M. J. and Schwarz, T. L. (2011a). PINK1 and Parkin target miro for phosphorylation and degradation to arrest mitochondrial motility. *Cell* **147**, 893-906. doi:10.1016/j.cell.2011.10.018
Wang, H., Song, P., Du, L., Tian, W., Yue, W., Liu, M., Li, D., Wang, B., Zhu, Y., Cao, C. et al. (2011b). Parkin ubiquitinates Drp1 for proteasome-dependent degradation: implication of dysregulated mitochondrial dynamics in Parkinson disease. *J. Biol. Chem.* **286**, 11649-11658. doi:10.1074/jbc.M110.144238
Wang, W., Xie, Q., Zhou, X., Yao, J., Zhu, X., Huang, P., Zhang, L., Wei, J., Xie, H., Zhou, L. et al. (2015). Mitofusin-2 triggers mitochondria Ca2+ influx from the endoplasmic reticulum to induce apoptosis in hepatocellular carcinoma cells. *Cancer Lett.* **358**, 47-58. doi:10.1016/j.canlet.2014.12.025
Wang, N., Wang, C., Zhao, H., He, Y., Lan, B., Sun, L. and Gao, Y. (2021). The MAMs structure and its role in cell death. *Cells* **10**, 657. doi:10.3390/cells10030657
Watanabe, S., Ilieva, H., Tamada, H., Nomura, H., Komine, O., Endo, F., Jin, S., Mancias, P., Kiyama, H. and Yamanaka, K. (2016). Mitochondria–associated membrane collapse is a common pathomechanism in SIGMAR 1 – and SOD 1 – linked ALS. *EMBO Mol. Med.* **8**, 1421-1437. doi:10.15252/emmm.201606403
White, C., Li, C., Yang, J., Petrenko, N. B., Madesh, M., Thompson, C. B. and Foskett, J. K. (2005). The endoplasmic reticulum gateway to apoptosis by Bcl-XL modulation of the InsP3R. *Nat. Cell Biol.* **7**, 1021-1028. doi:10.1038/ncb1302
Wu, H., Carvalho, P. and Voeltz, G. K. (2018). Here, there, and everywhere: the importance of ER membrane contact sites. *Science* **361**, eaan5835. doi:10.1126/science.aan5835
Wu, Y., Han, X., Su, Y., Glidewell, M., Daniels, J. S., Liu, J., Sengupta, T., Rey-Suarez, I., Fischer, R., Patel, A. et al. (2021). Multiview confocal super-resolution microscopy. *Nature* **600**, 279-284. doi:10.1038/s41586-021-04110-0
Yambire, K. F., Deus, C. M., Oliveira, P. J. and Raimundo, N. (2020). Mitochondria-lysosome crosstalk: from physiology to neurodegeneration | Elsevier enhanced reader. *Trends Mol. Med.* **26**, 71-88. doi:10.1016/j.molmed.2019.10.009
Yang, J., Vais, H., Gu, W. and Foskett, J. K. (2016). Biphasic regulation of InsP3 receptor gating by dual Ca2+ release channel BH3-like domains mediates Bcl-xL control of cell viability. *Proc. Natl. Acad. Sci. USA* **113**, E1953-E1962. doi:10.1073/pnas.1517935113
Yapa, N. M. B., Lisnyak, V., Reljic, B. and Ryan, M. T. (2021). Mitochondrial dynamics in health and disease. *FEBS Lett.* **595**, 1184-1204. doi:10.1002/1873-3468.14077
Yoshida, H., Matsui, T., Yamamoto, A., Okada, T. and Mori, K. (2001). XBP1 mRNA is induced by ATF6 and spliced by IRE1 in response to ER stress to produce a highly active transcription factor. *Cell* **107**, 881-891. doi:10.1016/S0092-8674(01)00611-0
Youle, R. J. and Strasser, A. (2008). The BCL-2 protein family: opposing activities that mediate cell death. *Nat. Rev. Mol. Cell Biol.* **9**, 47-59. doi:10.1038/nrm2308
Yu, W., Sun, Y., Guo, S. and Lu, B. (2011). The PINK1/Parkin pathway regulates mitochondrial dynamics and function in mammalian hippocampal and dopaminergic neurons. *Hum. Mol. Genet.* **20**, 3227-3240. doi:10.1093/hmg/ddr235
Zhou, J., Liu, C. Y., Back, S. H., Clark, R. L., Peisach, D., Xu, Z. and Kaufman, R. J. (2006). The crystal structure of human IRE1 luminal domain reveals a conserved dimerization interface required for activation of the unfolded protein response. *Proc. Natl. Acad. Sci. USA* **103**, 14343-14348. doi:10.1073/pnas.0606480103
Züchner, S., Mersianova, I. V., Muglia, M., Bissar-Tadmouri, N., Rochelle, J., Dadali, E. L., Zappia, M., Nelis, E., Patitucci, A., Senderek, J. et al. (2004). Mutations in the mitochondrial GTPase mitofusin 2 cause Charcot-Marie-Tooth neuropathy type 2A. *Nat. Genet.* **36**, 449-451. doi:10.1038/ng1341

Table S1. List of MAN-associated proteins modified in disease.

Disease	Protein involved	Modifications	Altered MAN	Refs.
PD	Parkin	LOF mutation leading to decrease in DRP-1 degradation	Mitochondrial fission	(Wang et al., 2011; Yu et al., 2011)
	Miro1	LOF mutation leading to impaired calcium homeostasis and increased mitophagy	Calcium signaling	(Grossmann et al., 2020)
	GBA1	LOF mutation abnormally prolonging lysosomal-mitochondrial tethering	Mitochondrial fission	(Kim et al., 2021)
Cancer	DRP-1	Increased activity leading to cell proliferation and migration	Mitochondrial fission	(Kashatus, 2018)
CMT-2A	MFN2	Mutation promoting mitochondrial hyperfusion by increased DRP-1 degradation	Mitochondrial fission	(Cartoni and Martinou, 2009)
	DRP-1	Increased R364W-MFN2-mediated ubiquitination leading to mitochondrial hyperfusion	Mitochondrial fission	(Das et al., 2021)
ALS	Sig-1R	LOF mutation leading to impaired binding to IP3R and mitochondrial dysfunction	Calcium signaling	(Al-Saif et al., 2011; Watanabe et al., 2016)
NASH and cancer	MFN2	Low expression leading to impaired PS transfer	Lipid exchange	(Hernández-Alvarez et al., 2019)
GM1-gangliosidosis	GLB1	LOF mutation leading to lysosomal dysfunction and neurodegeneration	Calcium signaling and Lipid exchange	(Sano et al., 2009)
NPC	NPC1	LOF mutation leading to accumulation of cholesterol at lysosomes causing cell death	Lipid exchange	(Wheeler and Sillence, 2020)

Abbreviations: amyotrophic lateral sclerosis (ALS); Charcot-Marie-Tooth disease type2A (CMT-2A); dynamin related protein 1 (DRP-1); β-glucocerebrosidase (GBA1); β-galactosidase (GLB1); Loss-of-function (LOF); mitofusin 2 (MFN2); Non-alcoholic steatohepatitis (NASH); Niemann-Pick type C (NPC); NPC intracellular cholesterol transporter 1 (NPC1); Parkinson's disease (PD); phosphatidylserine (PS); sigma-1R (Sig-1R).

References:

Al-Saif, A., Al-Mohanna, F. and Bohlega, S. (2011). A mutation in sigma-1 receptor causes juvenile amyotrophic lateral sclerosis. *Annals of Neurology* **70**, 913–919.

Cartoni, R. and Martinou, J.-C. (2009). Role of mitofusin 2 mutations in the physiopathology of Charcot–Marie–Tooth disease type 2A. *Experimental Neurology* **218**, 268–273.

Das, R., Kamal, I. M., Das, S., Chakrabarti, S. and Chakrabarti, O. (2021). MITOL-mediated DRP1 ubiquitylation and degradation promotes mitochondrial hyperfusion in CMT2A-linked MFN2 mutant. *Journal of Cell Science* jcs.257808.

Grossmann, D., Berenguer-Escuder, C., Chemla, A., Arena, G. and Krüger, R. (2020). The Emerging Role of RHOT1/Miro1 in the Pathogenesis of Parkinson's Disease. *Front Neurol* **11**, 587.

Hernández-Alvarez, M. I., Sebastián, D., Vives, S., Ivanova, S., Bartoccioni, P., Kakimoto, P., Plana, N., Veiga, S. R., Hernández, V., Vasconcelos, N., et al. (2019). Deficient Endoplasmic Reticulum-Mitochondrial Phosphatidylserine Transfer Causes Liver Disease. *Cell* **177**, 881-895.e17.

Kashatus, D. F. (2018). The Regulation of Tumor Cell Physiology by Mitochondrial Dynamics. *Biochem Biophys Res Commun* **500**, 9–16.

Kim, S., Wong, Y. C., Gao, F. and Krainc, D. (2021). Dysregulation of mitochondria-lysosome contacts by GBA1 dysfunction in dopaminergic neuronal models of Parkinson's disease. *Nat Commun* **12**, 1807.

Sano, R., Annunziata, I., Patterson, A., Moshiach, S., Gomero, E., Opferman, J., Forte, M. and d'Azzo, A. (2009). GM1-Ganglioside Accumulation at the Mitochondria-Associated ER Membranes Links ER Stress to Ca2+-Dependent Mitochondrial Apoptosis. *Molecular Cell* **36**, 500–511.

Wang, X., Winter, D., Ashrafi, G., Schlehe, J., Wong, Y. L., Selkoe, D., Rice, S., Steen, J., Lavoie, M. J. and Schwarz, T. L. (2011). PINK1 and Parkin target miro for phosphorylation and degradation to arrest mitochondrial motility. *Cell* **147**, 893–906.

Watanabe, S., Ilieva, H., Tamada, H., Nomura, H., Komine, O., Endo, F., Jin, S., Mancias, P., Kiyama, H. and Yamanaka, K. (2016). Mitochondria-associated membrane collapse is a common pathomechanism in SIGMAR 1 - and SOD 1 -linked ALS . *EMBO Molecular Medicine* **8**, 1421–1437.

Wheeler, S. and Sillence, D. J. (2020). Niemann–Pick type C disease: cellular pathology and pharmacotherapy. *Journal of Neurochemistry* **153**, 674–692.

Yu, W., Sun, Y., Guo, S. and Lu, B. (2011). The PINK1/Parkin pathway regulates mitochondrial dynamics and function in mammalian hippocampal and dopaminergic neurons. *Human Molecular Genetics* **20**, 3227–3240.