The ER-mitochondria interface, where Ca^{2+} and cell death meet

Ian de Ridder, Martijn Kerkhofs, Fernanda O. Lemos, Jens Loncke, Geert Bultynck*, Jan B. Parys*

KU Leuven, Laboratory for Molecular and Cellular Signaling, Department of Cellular and Molecular Medicine & Leuven Kanker Instituut, Campus Gasthuisberg O/N-1 B-802,
Herestraat 49, BE-3000 Leuven, Belgium

* Joint last authors, to whom correspondence should be addressed at geert.bultynck@kuleuven.be and jan.parys@kuleuven.be respectively

Keywords:

IP_3 receptor, mitochondria-associated membranes, ER-mitochondria tether, metabolism, apoptosis

Abstract

Endoplasmic reticulum (ER)-mitochondria contact sites are crucial to allow Ca^{2+} flux between them and a plethora of proteins participate in tethering both organelles together. Inositol 1,4,5-trisphosphate receptors (IP_3Rs) play a pivotal role at such contact sites, participating in both ER-mitochondria tethering and as Ca^{2+}-transport system that delivers Ca^{2+} from the ER towards mitochondria. At the ER-mitochondria contact sites, the IP_3Rs function as a multi-protein complex linked to the voltage-dependent anion channel 1 (VDAC1) in the outer mitochondrial membrane, via the chaperone glucose-regulated protein 75 (GRP75). This IP_3R-GRP75-VDAC1 complex supports the efficient transfer of Ca^{2+} from the ER into the mitochondrial intermembrane space, from which the Ca^{2+} ions can reach the mitochondrial matrix through the mitochondrial calcium uniporter. Under physiological conditions, basal Ca^{2+} oscillations deliver Ca^{2+} to the mitochondrial matrix, thereby stimulating mitochondrial oxidative metabolism. However, when mitochondrial Ca^{2+} overload occurs, the increase in $[Ca^{2+}]$ will induce the opening of the mitochondrial permeability transition pore, thereby provoking cell death. The IP_3R-GRP75-VDAC1 complex forms a hub for several other proteins that stabilize the complex and/or regulate the complex's ability to channel Ca^{2+} into the mitochondria. These proteins and their mechanisms of action are discussed in the present review with special attention for their role in pathological conditions and potential implication for therapeutic strategies.

1. Introduction

The endoplasmic reticulum (ER) is the largest intracellular Ca^{2+} store [1]. Ca^{2+} is actively transported into the ER by the sarco- and endoplasmic Ca^{2+} ATPases (SERCA) thereby reaching free intraluminal $[Ca^{2+}]_{ER}$ of up to 500 – 1000 µM, i.e. over 1000x higher than in the cytosol [2–4]. The steep Ca^{2+} gradient across the ER membrane serves as driving force for the flux of Ca^{2+} from the ER into the cytosol, thereby generating Ca^{2+} signals that propagate throughout the cell. Ca^{2+} flux from the ER is, depending on the cell type, mediated by inositol 1,4,5-trisphosphate (IP_3) receptors (IP_3R) [5–7], ryanodine receptors (RyR) [7–9] and/or members of the large group of Ca^{2+}-leak channels [10].

IP_3Rs form homo- and heterotetrameric Ca^{2+}-release channels in the ER membrane. They are encoded by three genes (*ITPR1*, *ITPR2*, and *ITPR3*) leading to three IP_3R isoforms (IP_3R1, IP_3R2 and IP_3R3, resp.) displaying a high level of homology [5,6]. These receptors are activated by IP_3 produced from phosphatidylinositol 4,5-bisphosphate after activation of cell-surface G protein-coupled receptors or receptors with tyrosine kinase activity. IP_3R opening and subsequent Ca^{2+} release only occur when all 4 subunits of the IP_3R channels are occupied by the IP_3 ligand [11]. The IP_3Rs vary in affinity for IP_3 [12] and their Ca^{2+}-flux properties are further regulated not only by local $[Ca^{2+}]$ and [ATP], but also by post-translational modifications such as phosphorylation and through interactions with regulatory and structural proteins [13–15].

The protein network around the IP_3R has important impacts on its function. Recently, K-Ras-induced actin-interacting protein (KRAP) was identified as a key protein that immobilizes IP_3Rs, thereby licensing them for the generation of Ca^{2+} puffs and global Ca^{2+} signals [16]. Other proteins can impact their Ca^{2+}-flux properties, such as in the case of Bcl-2 (inhibiting IP_3Rs [17]) or Bcl-XL (sensitizing IP_3Rs [18]), or their abundance, such as in the case of BRCA1-associated protein 1 (BAP1) (deubiquitinating and thus stabilizing IP_3Rs [19], F-box protein (FBXL2) and Erlin1/2 (ubiquitinating and thus tagging IP_3Rs for proteasomal degradation [20,21]). IP_3R abundance is not only controlled through accessory proteins but also through epigenetic factors, such as SMARCA2/4 that enhances chromatin accessibility to *ITPR3* and thus drives IP_3R3 expression [22]. Of note, some of these mechanisms may be isoform specific, whereby IP_3R1 is regulated by Erlin1/2 and IP_3R3 is controlled by BAP1, FBXL2 and SMARCA2/4. Furthermore, kinases can have profound effects on IP_3R function [13] with e.g. protein kinase A stimulating especially IP_3R1 [23] and protein kinase B (PKB/Akt) inhibiting the activity of all three IP_3R isoforms [24,25]. The resulting exquisite modulation of IP_3Rs enables the generation of complex spatio-temporal Ca^{2+} signals that control various aspects of cell behavior, including secretion, contraction, motility, gene transcription, metabolism, and cell fate in a cell type- and a condition-depending manner [26].

The regulation of cell fate by Ca^{2+} signaling is multi-faceted as it encompasses the processes of cell proliferation [27,28], differentiation [29–31], senescence [32–35], autophagy [28,36–40], as well as the various forms of cell death [28,36,39,41,42]. Please see also the further papers on the subject in this Special Issue.

Interestingly, at several locations in the cells, the ER forms close connections with the mitochondria. These contact sites harbor and/or control various processes, including Ca^{2+} and reactive oxygen species (ROS) signaling, lipid exchange, mitochondrial dynamics, inflammation, apoptosis and autophagy [43–50].

A particular aspect of intracellular Ca^{2+} signaling relevant in the framework of present review, is therefore the ability for the ER in general, and the IP_3R in particular, to provide at ER-mitochondria contact sites a quasi-synaptic Ca^{2+} flux towards the mitochondria [51]. Additionally, it should be mentioned that in cardiac and skeletal muscle cells, the RyR is also present at ER/sarcoplasmic reticulum (SR)-mitochondria contact sites, thereby providing efficient Ca^{2+} transfer towards the mitochondria [52–59]. Furthermore, ROS are produced by the mitochondria and the resulting H_2O_2 transients sensitize both the IP_3Rs and the RyRs located at the ER-mitochondria contact sites by redox modifications [60].

The mitochondrial Ca^{2+} uptake is mediated through the existence of membrane contact sites between these two organelles, Ca^{2+}-transport systems at the outer and the inner mitochondrial membrane (OMM and IMM resp.) and a strong negative potential across the IMM ($\Delta\psi$ of about -180 mV). Very recently, elegant work using synthetic IP_3R linkers that allow a rapamycin-controlled immobilization of IP_3Rs at the ER-mitochondria contacts convincingly demonstrated the unique role of IP_3R in enabling mitochondrial Ca^{2+} transfer and sustaining mitochondrial oxidative metabolism [61].

Mitochondria contain several Ca^{2+}-transport systems [62] of which the following two will be discussed in this review: the voltage-dependent anion channel (VDAC) that forms Ca^{2+}-permeable channels in the OMM [63,64] and the mitochondrial calcium uniporter (MCU) in the IMM. The latter is a low-affinity, high-capacity Ca^{2+}-selective ion channel that functions as part of a heteromeric protein complex. This MCU complex contains, besides four MCU subunits, a number of regulatory proteins (EMRE, MICU1, MICU2, MICU3, MCUb, and MCUR1). MICU1-3 are Ca^{2+}-binding proteins that control MCU opening. MICU1, an EF-hand intermembrane space (IMS) protein, controls the threshold and cooperativity of MCU's Ca^{2+}-transport function [65]. The endogenous ratio of MICU1/MCU varies across tissues, underlying tissue-specific MCU-dependent mitochondrial Ca^{2+} uptake phenotypes and downstream effects on oxidative metabolism [66]. EMRE appears essential

for MCU activity, while MCUb is a dominant-negative version of MCU, and its presence will decrease mitochondrial Ca^{2+} uptake. Finally, MCUR1 is most likely involved in the proper assembly of the MCU complex [67–70].

The close apposition between ER and mitochondria allowing efficient transfer of ER Ca^{2+} ions through the IP_3R to the mitochondrial matrix was first observed three decades ago by Rizzuto et al. [71,72]. Since then, the structure and physiological roles of ER-mitochondria contact sites have been amply studied. Several recent reviews have discussed various facets on these interorganellar contact sites and suggested their importance as therapeutic targets [44,73–77].

The properties and regulation of these contact sites and of the resulting Ca^{2+} flux from ER to mitochondria will be further discussed in sections 2 and 3. Furthermore, we will subsequently, discuss on the one hand the importance of $[Ca^{2+}]_{mit}$ for proper mitochondrial function (section 4) and on the other hand present a state-of-the-art view about the relation between high $[Ca^{2+}]_{mit}$ and apoptosis induction (section 5) before drawing conclusions (section 6).

2. Tethering of ER and mitochondria

Analysis of the ER-mitochondria contact sites by electron tomography indicated the existence of protein tethers with a length of 9-16 nm between smooth ER and the OMM and of 19-30 nm between rough ER and the OMM [78]. Moreover, using rapamycin-inducible linkers based on the interaction between FKBP12 and the FKBP12-rapamycin binding domain of mTOR, it was demonstrated that when the gap between ER and mitochondria was decreased below 7 nm, Ca^{2+} transfer was compromised, probably because the IP_3R could not be correctly accommodated in such a confined space [79]. Using longer linkers (15 nm), Ca^{2+} transfer occurred more efficiently. Within these contact sites, Ca^{2+} concentrations of 9 µM and higher were observed. These values correlate well with the Ca^{2+}-concentration range of 15-20 µM reported in an independent study [80].

The close apposition of the ER and mitochondria in contact sites is controlled by various components and dynamically regulated in response to changes in the cellular environment [81]. The contact between the ER surface of ER-mitochondria contact sites, termed mitochondria-associated ER membranes (MAMs) and the OMM is established by several tethering systems while anti-tethering systems ensure further modulation and spacing.

Historically, the first described tethering protein of ER-mitochondria contact sites was the cytosolic phosphofurin acidic cluster sorting protein 2 (PACS-2), a protein involved in vesicular sorting. Interestingly, PACS-2 depletion causes B-cell receptor-associated protein 31 (BAP31) cleavage at the ER, which leads to mitochondria fragmentation and uncoupling of the ER [82]. In fact, BAP31 couples

to FIS1, a mitochondrial fission protein located in the OMM (Figure 1), allowing for binding of pro-caspase 8. Stress signals can be transmitted from FIS1 to BAP31, facilitating cleavage of the latter to a pro-apoptotic p20 fragment, which elicits Ca^{2+} release from the ER and increased Ca^{2+} uptake in the mitochondria [83,84].

At ER-mitochondria contact sites, IP_3Rs reside in a macromolecular complex where the glucose-regulated protein 75 (GRP75) chaperone bridges the IP_3R to VDAC1 in the OMM [85] (Figure 1). All three IP_3R isoforms partake in ER-mitochondria contact, though IP_3R2 appears to be the most efficient for the delivery of Ca^{2+} ions to the mitochondria [86]. Moreover, the tethering function of the IP_3R does not dependent on its Ca^{2+}-release activity, since a pore-dead IP_3R mutant too can contribute to the formation of ER-mitochondria contacts [86]. Other proteins can associate with and recruit IP_3R to the MAMs, of which we will highlight some recent examples. At the functional level, various IP_3R isoforms may mediate ER-mitochondrial Ca^{2+} flux, depending on cell type, physiological conditions and/or cellular stress inducers. However, a prominent role has been implicated for IP_3R3 channels and their regulation in both sustaining cell bioenergetics [87] as well as mediating cell death [19,20].

A subpopulation of the ER stress sensor inositol-requiring enzyme 1 (IRE1α) resides in MAMs. Apart from its canonical role in the unfolded protein response (UPR), IRE1α can scaffold IP_3R in the MAMs [88]. Moreover, the MAM-enriched chaperone sigma-1 receptor (S1R) can stabilize IRE1α and enhance its endonuclease activity necessary for the UPR [89]. In the MAMs, S1R also stabilizes the IP_3R, in this way enhancing ER-mitochondria Ca^{2+} fluxes [90]. Moreover, the IP_3R-S1R complex is formed upon chronic ER stress, a feature that is enhanced by pharmacological S1R agonists. Besides IRE1α, also another UPR player, protein kinase RNA-like ER kinase (PERK), has been found to act as a structural tether that help stabilizing the ER-mitochondria interface, thereby ensuring the transfer of ROS and Ca^{2+} signals [91]. Moreover, PERK acts as a docking site for the recruitment of E-Syt1 to MAMs, thereby enabling the adequate transfer of phospholipids from ER to mitochondria to ensure mitochondrial respiration [92].

In the OMM of cardiomyocytes, FUN14 domain-containing 1 (FUNDC1), binds to IP_3R2 and is important for ER-mitochondria contact (Figure 1). Loss of FUNDC1 leads to abrogated ER-mitochondria contact and cardiac dysfunction in mice [93].

Mitofusin (Mfn) 2, a GTPase mediating mitochondrial fusion and dynamics, is the subject of some controversy. Mfn2 resides both at the ER and OMM, while Mfn1 is only present at the OMM. As such, ER-resident Mfn2 has been proposed as a ER-mitochondria tethering protein by forming interorganellar complexes with the OMM-resident Mfn2 (homotypic complexes) and Mfn1

(heterotypic complexes) (Figure 1) [94,95]. Other studies, however, reported an increase in ER-mitochondria contact sites upon Mfn2 downregulation [96–102], suggesting rather an "anti-tethering" function. For now, the exact role of Mfn2 as either a tethering or anti-tethering protein remains to be fully established and is further critically discussed in Filadi et al. [103]. Furthermore, Mfns are regulated by ubiquitination. MITOL, encoded by *MARCH5*, ubiquitinates mitochondrial Mfn2 [104]. Ubiquitination at K63 leads to Mfn2 oligomerization and MAM formation, while K192 poly-ubiquitination is necessary for GTP binding to Mfn2. Parkin, one of the proteins in which mutations are linked to Parkinson's disease (PD), is an E3 ubiquitin ligase targeting Mfn1 and Mfn2. Such ubiquitination may lead to Mfn degradation [105], but can also promote ER-mitochondria contact [106] via K416 ubiquitination of Mfn2 [107]. In addition, also trichoplein/mitostatin, a putative tumor suppressor, interacts with Mfn2 but thereby acts as a negative regulator of ER-mitochondria tethering [108].

Another pair of interacting proteins that is crucial for ER-mitochondrial contact is the ER-membrane residing vesicle-associated membrane protein-associated protein B (VAPB) and protein tyrosine phosphatase-interacting protein 51 (PTPIP51) at the OMM (Figure 1). In neuronal-like cells, the overexpression of Tar DNA-Binding Protein 43, a protein associated with amyotrophic lateral sclerosis and dementia, activated glycogen synthase kinase-3β (GSK3β) which in turn disrupted the VAPB-PTPIP51 complex and ultimately decreased ER-mitochondria contacts [109]. VAPB and PTPIP51 localize to neuronal synapses, where they stimulate ER-mitochondria contact and neuronal activity [110]. As such, pharmacologically targeting ER-mitochondria contact formation, e.g. by GSK3β inhibitors, is being considered for the treatment of neurodegenerative diseases [111].

Finally, in the search of a mammalian ortholog of the ER-mitochondria tethering complex in yeast, the ERMES complex, a protein called PDZ domain-containing protein 8 (PDZD8) was discovered [112], which contains a synaptotagmin-like mitochondrial lipid-binding protein (SMP) domain (Figure 1). Three out of the four ERMES proteins also contain such SMP domains. PDZD8 is a MAM-localized protein and positively influences ER-mitochondria contact, whereas its deficiency in dendrites of cortical neurons leads to impaired ER-mitochondria Ca^{2+} transfer [112]. Furthermore, together with Ras-related protein 7 and protrudin, PDZD8 localized to a tripartite contact site between the ER, late endosomes and mitochondria [113]. While the role of PDZD8 in contact site integrity is apparent, its mitochondrial target has not yet been identified.

In contrast to the above-mentioned tethers, also spacer proteins have emerged. For instance, fetal and adult testis expressed transcript (FATE) 1, a member of the mitochondrial fission factor family and cancer-testis antigen, act as an organellar uncoupler of ER and mitochondria [114,115]. In

adrenocortical carcinoma cells, FATE1-protein levels are upregulated via transcription factor SF-1. This leads to a decreased coupling between ER and mitochondria, thereby reducing mitochondrial Ca^{2+} uptake and subsequently the sensitivity to Ca^{2+}-dependent pro-apoptotic stimuli and apoptosis induced by chemotherapeutic drugs [114].

During the past two decades, several proteins that could be involved in ER-mitochondria tethering have been discovered. For now, a knowledge gap exists in the sense that it is unclear how the different tethering proteins cooperate in establishing, maintaining and controlling ER-mitochondria contacts. Presumably, the discovery of further structural or regulating actors participating in the apposition of both organelles and a better understanding of their dynamics will allow to paint a more complete picture. While some ubiquitous proteins involved in the tethering of ER to mitochondria are likely mediating ER-mitochondria contact sites in cells of virtually all tissues, such as the IP_3R and IRE1α, others are tissue specific, such as FUNDC1 in cardiomyocytes. Moreover, the MAMs composition is also dynamically regulated dependent on cell types, physiological demands and/or cell-stress conditions. Recently, it became clear that IP_3Rs are motile and can travel in and out ER-mitochondria contact sites [61]. When IP_3Rs are trapped at such contact sites, they enable ER-mitochondria Ca^{2+} transfer. In addition to this, an increasing number of kinases and phosphatases have been identified to reside and act at the MAMs thereby impacting local protein function, protein localization and protein interaction networks [116]. The multitude of tethering proteins, the differences among cell types and mechanisms ensuring dynamic control underscore the necessity for cells to very precisely tailor the ER-mitochondria contacts according to their needs.

3. The IP_3R as Ca^{2+}-release channel and tether

3.1. The IP_3R-GRP75-VDAC1 core complex

As already indicated above (section 2), IP_3Rs physically connect to the mitochondrial VDAC1 channels through tethering via GRP75 [85]. GRP75 is a molecular chaperone that belongs to the heat shock protein 70 family and is mainly localized in the mitochondrial matrix [117,118]. Additionally, GRP75 heavily influences the Ca^{2+} signaling related to its tethering function at the MAMs, enabling the quasi-synaptic Ca^{2+} transfer from the ER into the mitochondria [85]. The MCU complex in the IMM is proposed to also reside in the proximity of the IP_3R-GRP75-VDAC1 tethering complex (Figure 2), and these four proteins form the Ca^{2+}-transporting axis, even though the MCU is not known to directly interact with any of the other proteins within the axis [119,120]. The physical linkage between ER and mitochondria in general and in particular between IP_3Rs and VDAC1 through GRP75 allows the formation of high [Ca^{2+}] microdomains at the MAMs, which helps to surmount the low Ca^{2+} affinity of the MCU complex [79,80]. This efficient transfer of Ca^{2+} to the mitochondrial matrix is however also

due to the positive cooperativity enabled by the presence of MICU1 and MICU2 within the MCU complex [65].

The IP_3R-GRP75-VDAC1 complex at the MAMs functionally impacts cellular energetics (see section 4) as well as cell death (see section 5). Dysregulation of the tethering complex, for example through silencing or deletion of GRP75, reduces Ca^{2+} flux from the ER to the mitochondria and will result in avoidance of the cell death that would result from mitochondrial Ca^{2+} overload [121–124]. It should however be pointed out that recent results [61] suggested that VDAC1 is present at such a high level in the OMM, that its presence in the tether may be dispensable and that IP_3Rs could also be tethered to the mitochondria via other proteins, e.g. TOM70, without impeding the Ca^{2+} transfer. TOM70, a translocase located at the OMM, interacts with IP_3Rs thereby supporting their functional recruitment in close proximity of the mitochondria, enabling ER-mitochondria Ca^{2+} transfer and sustaining mitochondrial bio-energetics [87].

3.2. Additional interaction partners of the IP_3R-GRP75-VDAC1 complex

In recent years, an increasing number of proteins have emerged that modulate the formation and functionality of the IP_3R-GRP75-VDAC1 complex (Table 1). Here we will discuss the most important proteins and their features, thereby focusing on the most recent discoveries.

DJ-1, a MAM-localized protein related to early onset PD, is one of the IP_3R-GRP75-VDAC1 interaction partners that received a lot of attention in recent years (Figure 2). DJ-1 physically interacts with the IP_3R3-GRP75-VDAC1 complex, thereby maintaining MAM integrity and functionality. Consequently, loss of DJ-1 in M17 neuroblastoma cells results not only in the disruption of the IP_3R-GRP75-VDAC1 complex itself but also of the MAMs [125–127]. Interestingly, the MAMs could be restored by wild-type DJ-1 but not by DJ-1 mutants linked to PD. Such DJ-1 disease mutants were impaired in interacting with the IP_3R3-GRP75-VDAC1 tethering complex [125]. Moreover, DJ-1 influences IP_3R3 turnover and aggregation, with decreased DJ-1 levels associated with less complex formation and less Ca^{2+}-induced IP_3R3 degradation, leading to accumulation of the latter at the MAMs [125]. Mutant DJ-1 could therefore be at the root of the malfunctioning ER-mitochondria communication observed in PD and could thus form a potential therapeutic target.

Another protein targeting the IP_3R-GRP75-VDAC1 tethering complex is transglutaminase type 2 (TG2) (Figure 2), which mediates mitochondrial homeostasis in stress conditions among numerous other functions [117]. TG2 interacts with VDAC1 and GRP75 thereby hampering its interaction with IP_3R3. This results in reduced ER-mitochondria Ca^{2+} flux, a decreased number of MAMs, and modified levels of MAM proteins. Remarkably, it is believed that excessive IP_3R3-GRP75 interaction in the absence of TG2, reduce the functionality of the IP_3R-GRP75-VDAC1 complex at the MAMs. Hence, TG2

modulates the amount of GRP75 present in the tethering complex. Interestingly, TG2 also decreases Ca^{2+} flux through IP_3R1 via posttranslational modification of Gln2746, thereby locking the receptor in its closed conformation [128]. Yet, it is unknown whether this TG2-induced modification also impacts IP_3R1's tethering function at the MAMs.

Recently, α-synuclein was also identified as an IP_3R-GRP75-VDAC1 complex regulator in the context of PD [129]. Overexpression of α-synuclein impairs IP_3R-GRP75 interaction as well as decreases mitochondrial Ca^{2+} overload and the number of MAMs without affecting DJ-1 levels. Nonetheless, a direct interaction between α-synuclein and any of the IP_3R-GRP75-VDAC1 complex partners is yet to be determined.

In addition to this, also Wolfram syndrome 1 (WFS1), whose loss of function leads to Wolfram syndrome, has been identified as a MAM-resident protein affecting local IP_3R function [130]. At the MAMs, WFS1 forms a complex with IP_3Rs, likely through the neuronal calcium sensor-1 (NCS-1), thereby supporting the IP_3R-VDAC1 axis and sustaining IP_3R-mediated ER-mitochondria Ca^{2+} transfer [131]. NCS-1 is a known IP_3R-binding partner that enhances IP_3R-mediated Ca^{2+} release [132]. However, NCS-1 is also susceptible to degradation, particularly in conditions of sustained $[Ca^{2+}]_{cyt}$ elevations [133]. Loss of WFS1 disturbs MAM formation and impairs mitochondrial bio-energetics. At the molecular level, lack of WFS1 results in the degradation of the NCS-1 protein, thereby impairing IP_3R function and reducing ER-mitochondria Ca^{2+} transfer [134]. How WFS1 stabilizes NCS-1 is not fully understood, but NCS-1 degradation in WFS1-deficient cell models could be due to loss of physical interactions between WFS1 and NCS-1 or be a consequence of sustained ER stress and UPR signaling.

However, NCS-1 is not easy to target pharmacologically. Therefore, more recent work focused on more therapeutically tangible targets such as the S1R. This protein resides at the ER and particularly the MAMs, supports ER-mitochondrial Ca^{2+} transfer and can be stimulated by chemical S1R agonists [135]. Fascinatingly, pharmacological S1R activation using the agonist PRE-084 can restore MAM function and ER-mitochondria Ca^{2+} transfer in a WFS1 knockout disease model, thereby correcting several mitochondrial defects such as compromised mitochondrial bio-energetics and elevated autophagy and mitophagy. These improvements at the subcellular level also translated at the behavioral level in studies performed in WFS1-deficient zebrafish and mouse models. This further underpins the interconnection of different MAM-resident proteins and their ability to serve as targets to correct or compensate (here NCS-1 or S1R) for deficiencies caused by other MAM-resident proteins (here WFS1). However, whether S1R stimulation or overexpression/stimulation of other

MAM components would be able to rescue other deficiencies in MAM tethers or MAM-resident proteins remains unknown but are interesting avenues to explore further.

The IP_3R-GRP75-VDAC1 complex therefore functions as a hub for a variety of proteins controlling in this way ER-mitochondria Ca^{2+} flux and downstream pathways, including mitochondrial energetics and cell survival versus cell death. Not only does this intricate web of protein modulation has major implications for general cell regulation, the IP_3R-GRP75-VDAC1 complex and its regulatory effects have also been revealed to be heavily dysregulated in a plethora of diseases, including cancers, neurodegenerative diseases, and diabetes mellitus.

3.3.The IP_3R-GRP75-VDAC1 complex in patho(physio)logical situations

In neuronal cells, the Ca^{2+}-mediated communication between the ER and mitochondria seems to be extensively modulated in many neurodegenerative disorders, with effects at both the structural and the functional level of the MAMs. For example, in Alzheimer's disease (AD), elevated Ca^{2+} flux from the ER to the mitochondria was observed along with increased MAM formation and elevated levels of IP_3Rs and VDAC1, which were all linked to an increased level of amyloid β (Aβ) [136–138]. The excessive mitochondrial Ca^{2+} loading was also observed in the neurons of living brain by performing multiphoton microscopy via a cranial window on mouse models for AD [139]. Interestingly, several MAM proteins, such as PACS-2 and S1R, were also found to be upregulated at the MAMs in both AD mouse models and AD patient cells, thereby modulating the IP_3R-GRP75-VDAC1 complex [137]. In the context of truncated apolipoprotein E4's neurotoxicity, upregulation of GRP75-protein levels were also observed in AD cell models with clear ties to its functioning at the MAMs [122]. Consistent with this, reducing ER-mitochondria contact by decreasing the protein levels of the tethering protein PDZD8 exerts a neuroprotective effect and sustains locomotor function in a Drosophila model of AD based on Aβ42 expression, though whether this also entailed reduced mitochondrial Ca^{2+} uptake was not assessed [140].

In recent years, dysregulation of the IP_3R-GRP75-VDAC1 complex further emerged as a critical component underlying the pathogenesis of other neurodegenerative diseases besides AD, including PD, Huntington's disease, motor neuron diseases, and amyotrophic lateral sclerosis. A more extensive view on the involvement of MAM-localized Ca^{2+} signaling in neurons and in neurodegeneration was recently reviewed [141].

The IP_3R-GRP75-VDAC1 complex is also of utmost importance in skeletal muscle cells, which is not surprising, considering the high energy consumption of muscle cells and the key role of mitochondrial Ca^{2+} homeostasis on mitochondrial bioenergetics. A reduced number of MAMs and defective mitochondrial function, often directly linked with defects at the level of the IP_3R-GRP75-VDAC1

complex, have been implicated in muscle pathophysiology related to defective skeletal muscle functioning, such as insulin resistance [142,143], ageing [144–146], and muscle dystrophy [147]. In the latter case, MAMs architecture and their capacity for mitochondrial Ca^{2+} transfer was diminished. In mice suffering from age-related muscle dystrophy, the number of mitochondria connected with the RyR was decreased, provoking thus a reduction in contact sites between ER/SR and mitochondria [144]. Clear relations were observed between muscle dystrophy and the expression level of IP_3R, GRP75, and VDAC1 [145], MCU [148] as well as Mfn2 [144]. These findings underscore the importance of MAMs and their Ca^{2+}-transport function in muscle cells. For a more extensive view on the topic, we refer to Zhang et al. [149].

Furthermore, GRP75-controlled ER-mitochondria Ca^{2+} flux appears to play a central role in the pathogenesis of asthma, thereby evoking excessive ROS production and cytokine release in imbalanced T-helper 1 and 2 response [150]. In the context of cisplatin resistance of ovarian cancers, increased MAM formation was observed as a result of GRP75 and VDAC1 overexpression [151]. This phenotype is thought to overcome the mitochondrial dysfunction induced by cisplatin as well as the resulting apoptosis.

Finally, in hepatocytes, ER-mitochondria Ca^{2+} communication is important for adequate responses to insulin and glucose homeostasis [115]. Decreased ER-mitochondria contacts is an early event in diet-induced obesity in mice and is also observed in liver biopsies of obese patients with type 2 diabetes. Interestingly, changing ER-mitochondrial contact is sufficient to impact the health and function of hepatocytes. On the one hand, disrupting ER-mitochondrial contact, e.g. by forced expression of the spacer protein FATE1, is sufficient to impair the physiological function of hepatocytes, eventually resulting in hepatic steatosis. On the other hand, artificially restoring ER-mitochondria contacts through synthetic linkers is able to prevent diet-induced glucose intolerance.

Taken together, these data underscore the importance of proper distance between ER and mitochondria in general and of GRP75 as member of a complex with the IP_3R and VDAC1 in particular, in stress responses, redox and Ca^{2+} homeostasis. Moreover, these findings indicate the potential of the IP_3R-GRP75-VDAC1 complex and its modulating partners as therapeutic targets in a wide range of diseases.

4. Physiological functions of mitochondrial Ca^{2+}

Mitochondrial Ca^{2+} uptake is a finely controlled process indispensable for regulating cell metabolism and cell fate. In physiological conditions, increases in $[Ca^{2+}]_{cyt}$ caused by different energy-requiring events, such as stimulation by hormones, secretion, muscle contraction, can be transmitted to the mitochondria in order to upregulate ATP supply and keep up with energy consumption [100,152].

Several events in the mitochondria, from enzyme activities to substrate transporters, can be activated by mitochondrial Ca^{2+} in order to stimulate aerobic metabolism [100,152,153].

Mitochondrial Ca^{2+} stimulates energy production by stimulating the activity of FAD-linked glycerol phosphate dehydrogenase (GPDH), pyruvate dehydrogenase (PDH), NAD-isocitrate dehydrogenase (IDH) and oxoglutarate dehydrogenase (OGDH) [153]. Moreover, by specifically capturing IP_3Rs at the contact sites between ER and mitochondria, Katona et al. could elegantly demonstrate that the resulting increase in $[Ca^{2+}]_{mit}$ led to activation of these dehydrogenases [61]. In the absence of IP_3Rs, mitochondrial bioenergetics is compromised and autophagy is activated [154].

GPDH is a Ca^{2+}-sensitive enzyme involved in the glycerol phosphate shuttle, catalyzing the oxidation of cytosolic glycerol-3-phosphate to dihydroxyacetone phosphate. Electrons from this reaction are transferred via FAD to the electron transport chain (ETC). GPDH is localized in the IMM, with the binding sites for glycerol phosphate and Ca^{2+} facing the IMS, being therefore readily accessible to Ca^{2+} and substrates from the cytosol [155,156]. $[Ca^{2+}]$ in the micromolar range activates GPDH by lowering the K_m for glycerol phosphate [155–157].

PDH is a dehydrogenase at the beginning of the tricarboxylic acid (TCA) cycle that catalyzes the conversion of pyruvate to acetyl-CoA, an irreversible step in the carbohydrate metabolism [158]. PDH is a macro complex localized in the mitochondrial matrix, formed by multiple copies of pyruvate decarboxylase, dihydrolipoate dehydrogenase, and dihydrolipoate acetyltransferase [159]. The amount of the products acetyl-CoA and NADH in relation to the substrates CoA and NAD^+, and the phosphorylation/dephosphorylation of pyruvate decarboxylase subunits are ways to strictly regulate PDH activity [158], and the latter step is regulated by Ca^{2+}. The phosphorylation of PDH by pyruvate dehydrogenase kinases (PDKs) causes PDH inhibition, which is specifically reversed by the Mg^{2+}- and Ca^{2+}-dependent pyruvate dehydrogenase phosphatase isoform 1 (PDP1) [160–162]. Ca^{2+} thereby binds with a K_d of 1 µM to the complex formed by PDP1 and dihydrolipoate acetyltransferase and stimulates the dephosphorylation of pyruvate decarboxylase [160,161].

In contrast, the conversion of isocitrate to α-ketoglutarate is directly controlled by mitochondrial Ca^{2+}. This chemical reaction is catalyzed by IDH, a 320 kDa macromolecular complex formed by three different subunits [163,164]. The enzymatic activity of IDH is inhibited by increasing ATP/ADP and NADH/NAD^+ ratios. Lower ATP/ADP ratios promotes the interaction of Ca^{2+} to IDH, which decreases its K_m for isocitrate, facilitating its activation [165–167]. The dissociation constants for Ca^{2+} activation of IDH range between 5 and 50 µM, depending on the ATP/ADP ratio [165,166].

The last TCA cycle dehydrogenase controlled by mitochondrial Ca^{2+} levels is OGDH, a multi-enzyme complex formed by dihydrolipoamide succinyl transferase, 2-oxoglutarate decarboxylase and

dihydrolipoamide dehydrogenase. OGDH is responsible for the conversion of α-ketoglutarate to succinyl-CoA. OGDH activity is inhibited by increases in the concentrations of the metabolites succinyl-CoA and NADH. On contrary, increases of [Ca^{2+}] and lower ATP/ADP ratios raise OGDH activity [168]. The decrease of ATP/ADP ratio remarkably sensitizes OGDH to Ca^{2+}, while Ca^{2+} directly binds and reduces the K_m for oxoglutarate [165,168,169]. Therefore, the K_m for oxoglutarate varies from 0.2 to 2 μM depending to the presence of ADP [165], and the dissociation constant for Ca^{2+} binding ranges between 1 and 7 μM [166]. The 2-oxoglutarate decarboxylase contains a regulatory Ca^{2+}-binding site, which contributes to the Ca^{2+}-sensitivity of the OGDH complex [169–171].

In addition to the stimulatory effect of Ca^{2+} on the dehydrogenases named above, Ca^{2+} can directly activate the ETC and the F_1F_o ATP synthase. Ca^{2+} at low micromolar concentration significantly increases the H^+ conductance of complexes I, III and IV of the ETC, doubling the maximum velocity of oxidative phosphorylation [172]. Regarding the F_1F_o ATP synthase, which is the complex responsible for producing ATP using the H^+ gradient generated by the ETC, the increase of Ca^{2+} in the mitochondrial matrix can directly activate this enzyme [173].

Finally, Ca^{2+} regulates the shuttles of different nucleotides, metabolites and cofactors across the IMM by regulating the Ca^{2+}-binding mitochondrial carriers (CaMCs) located in this membrane. Based on the number of amino acids and the mitochondrial carrier homology sequences, the CaMCs family can be divided in longer (L-CaMCs) and shorter (S-CaMCs) members [100].

L-CaMCs, also known as aralar1 and citrin, are two isoforms of the aspartate/glutamate carrier (AGC) involved in the malate-aspartate NADH shuttle (MAS) and the urea cycle. L-CaMCs function as an electrogenic exchange of mitochondrial aspartate for cytosolic glutamate and a H^+, which contributes to the entry of NADH into mitochondria and the mitochondrial metabolism in a Ca^{2+}-dependent manner [174–177]. Low [Ca^{2+}] (nanomolar range) already activate AGC due to the presence of EF-hand motifs located in the N-terminal region, facing the IMS. In tissues in which the main expressed isoform is aralar1, e.g. brain, half-maximal activation occurs at ~300 nM Ca^{2+} [178], while in liver, in which citrin is the main isoform, half-maximal activation occurs at ~150 nM Ca^{2+} [179].

S-CaMCs, as ATP-Mg^{2+}/Pi carriers, regulate the level of adenine nucleotides in the mitochondrial matrix by exchanging across the IMM ADP or ATP bound to Mg^{2+} for one inorganic phosphate, directly impacting cell metabolism [180]. The activity of S-CaMCs is strictly Ca^{2+}-dependent at micromolar concentration due to the presence of a calmodulin-like motif at the N-terminus of the S-CaMCs, facing the IMS [181,182]. The hypothesis is that the calmodulin-like motif controls an auto-inhibitory region in the N-terminus of the S-CaMCs: in the absence of Ca^{2+} this auto-inhibitory region

blocks the IMS side of the carriers, while the presence of Ca^{2+} promotes conformational changes in the N-terminus and removes this blockage, allowing transport of the adenine nucleotides [183–185].

Apart from triggering autophagy [38,186], a decreased Ca^{2+} transfer from ER to mitochondria can also influence mitochondria in other ways. $[Ca^{2+}]_{mit}$ can affect both the morphology and the intracellular localization of the mitochondria [187]. In cancer cell lines, mitochondrial Ca^{2+} overload stimulated fragmentation of the mitochondria, while low $[Ca^{2+}]_{mit}$ led to hyperfusion, characterized by the appearance of elongated, highly interconnected mitochondria [188]. Furthermore, also the intracellular mobility of mitochondria can be affected by $[Ca^{2+}]_{mit}$ [189]. The average speed of mitochondria in the axons of hippocampal neurons ranged from 0.1 to 0.6 µm/sec that inversely correlated with the $[Ca^{2+}]_{mit}$. A lower $[Ca^{2+}]_{mit}$ thus led to a faster movement of the mitochondria, potentially ensuring redistribution to a different intracellular location [189]. Moreover, the same study found that mutations in the EF-hand domain of Miro1, a Ras GTPase implicated in mitochondrial mobility, reduced mitochondrial Ca^{2+} influx, further supporting a role of $[Ca^{2+}]_{mit}$ in the control of mitochondrial mobility.

5. Mitochondrial Ca^{2+} overload and apoptosis

It is important to realize that mitochondrial Ca^{2+} uptake functions as a double-edged sword. Although mitochondrial Ca^{2+} uptake is essential for cellular energy homeostasis and survival (see section 4), excessive ER-mitochondrial Ca^{2+} transfer can trigger cell death [190]. While Ca^{2+} signaling affects multiple cell death modalities through various pathways [190,191] they do not all necessarily involve mitochondria, e.g. calpains can be directly activated by increasing $[Ca^{2+}]_{cyt}$ [192]. For the purpose of this review, we will focus on the relation between ER-mitochondria Ca^{2+} transfer and cell death.

5.1. Relation between $[Ca^{2+}]_{mit}$ and the occurrence of apoptosis.

It has long been recognized that excessive Ca^{2+} uptake into the mitochondria is a trigger for cellular demise [190]. This is due to the fact that Ca^{2+} acts as a trigger for mitochondrial permeability transition pore (mPTP) opening [193–195]. At the molecular level, Ca^{2+} accumulation in the mitochondrial matrix evokes the disassembly of complex II of the ETC. Enzymatically active components of this complex are released from the IMM-resident partners, thereby generating excessive ROS in an uncontrolled way. The culprit for the disassembly of complex II is the loss of cardiolipin, a Ca^{2+}-binding IMM lipid that upon Ca^{2+} binding dissociates from complex II [196]. The mPTP opening signifies a non-selective permeabilization of the IMM, which leads to an osmotic imbalance [197,198]. This, in turn, leads to influx of water, OMM permeabilization and the release of cytochrome c and other pro-apoptotic factors [199]. These factors trigger apoptosome assembly and downstream activation of caspases, the executioners of apoptosis. It is important to note that mPTP

opening is not exclusively linked with apoptotic cell death but may also be associated with necrosis [190,191].

From a molecular point of view, mPTP is a Ca^{2+}-sensitive pore in the IMM, the molecular identity of which is still heavily debated [197,200]. Throughout the last decades, multiple candidates, such as VDAC1, cyclophilin D (CyPD) and the adenine nucleotide translocator (ANT) have been proposed to constitute the mPTP, but so far, none have withstood the scrutiny of robust genetic testing. The latest findings are therefore summarized below.

The most recent addition to the list of mPTP candidates is the F_1F_o ATP synthase [194,201–204]. Indeed, the F_1F_o ATP synthase interacts with a number of previously identified regulators of mPTP opening, such as CyPD [205] while factors that control opening of the mPTP, such as ATP, ADP and inorganic phosphate, closely correlate with the biological function of the F_1F_o ATP synthase [197]. In addition, the Bernardi group demonstrated that Ca^{2+} binding to the β-subunit of the F_1F_o ATP synthase is able to induce conformational changes in the protein [194]. In addition, the mitochondrial chaperone TRAP1 is able to interact with and modulate the function of the F_1F_o ATP synthase, while it counteracts CyPD-mediated facilitation of the mPTP [206]. Currently, two models exist for the F_1F_o ATP synthase to function as the mPTP. The first proposes the membrane-embedded c-ring subunit of the F_1F_o ATP synthase to function as the mPTP [201,203,204], the second puts forward that dimerization of the F_1F_o ATP synthase is the pore-forming mechanism of the mPTP [194,202]. While the knockout of the c subunit of the F_1F_o ATP synthase does not abolish mPTP opening, the peak conductance of the mPTP was significantly lower in cells lacking the c subunit [207]. Also the dimerization model has been disputed by studies showing that monomeric forms of the F_1F_o ATP synthase are still permissive for mPTP opening [208].

Interestingly, recently, a dual model for mPTP opening has been proposed by the Molkentin lab [209]. They revisited the rejected hypothesis that the earlier proposed ANT would be responsible for mPTP opening and generated a knockout model where all three murine isoforms of the ANT are absent. In this model, they found that simultaneous knockout of CyPD or its pharmacological inhibition, abolished mPTP opening. Their data suggest that there are two pore components, one ANT-dependent and one CyPD-dependent that requires another molecular player [209]. This dual model for mPTP is also supported by work from the Pavlov group, indicating that deficiency in the c-subunit of the F_1F_o ATP synthase causes loss of mPTP opening, while at the same time CyPD-dependent channel activity remains [210]. In conclusion, the molecular identity of the mPTP remains up for debate.

5.2. Regulation of Ca^{2+}-mediated apoptosis

The control of Ca^{2+}-mediated apoptosis is complex and can be regulated at various levels: IP_3R-mediated Ca^{2+} release from the ER, Ca^{2+} uptake in the mitochondria through VDAC1 and MCU and the distance between ER and mitochondria at the locations where they are closely juxtaposed. IP_3R-mediated Ca^{2+} release is controlled in a variety of ways. Firstly, IP_3R expression levels are important determinants of IP_3R-mediated Ca^{2+} release. Thereby, it is crucial to remember that the IP_3R exists in three different isoforms that in spite of their homology are endowed with different Ca^{2+}-signaling properties [5,211–214]. It was thought for a long time that IP_3R3 was preferentially responsible for the transfer of pro-apoptotic Ca^{2+} signals to the mitochondria [215,216]. Recently, however, this model has been challenged with new evidence that also IP_3R1 and IP_3R2 are able to provoke pro-apoptotic Ca^{2+} signaling [211,213,217] with IP_3R2 even being the most efficient [86].

While the combination of IP_3R expression levels and isoforms already allows for the fine-tuning of ER-mitochondria Ca^{2+} transfer, Ca^{2+} signals can be further controlled through post-translational modifications. For example, PKB/Akt-mediated phosphorylation of IP_3R3 decreases its Ca^{2+}-flux properties and thus protects cells against mitochondrial Ca^{2+} overload and apoptosis [24,216]. IP_3R-mediated Ca^{2+}-release is actively disinhibited by the phosphatase and tensin homolog (PTEN) [218], which dephosphorylates IP_3R3. Moreover, PTEN also displaces FBXL2 from its binding site on the IP_3R3, stabilizing the level of the latter, thereby increasing pro-apoptotic Ca^{2+} signaling to the mitochondria [20]. These mechanisms become important in cancer cells in which PKB/Akt functions as an oncogene and PTEN as a tumor suppressor. Cancer cells often lose PTEN at the genetic level or acquire loss-of-function mutations resulting in a dysfunctional PTEN protein [219]. This skews the balance in favor of PKB/Akt-mediated phosphorylation of IP_3Rs and suppression of ER-mitochondria Ca^{2+} transfer. This in turn, protects cancer cells against mitochondrial Ca^{2+} overload and apoptosis, at least in vitro [211,218].

Thirdly, IP_3Rs are functionally modulated by their binding partners. More than a hundred proteins have been identified to interact with IP_3Rs [14], including many known oncogenes and tumor suppressors that affect IP_3R activity and consequently cell death or cell survival [220,221]. However, also other proteins can similarly affect IP_3R function, including metabolic enzymes as hexokinase 2 [222] and pyruvate kinase M2 [223].

For the purpose of this review, we will limit ourselves to the discussion of the Bcl-2-protein family as the latter family plays a crucial role in the regulation of canonical apoptosis by controlling mitochondrial outer membrane permeabilization (MOMP) [224,225]. MOMP is executed by the multi-BH-domain-containing members of the Bcl-2 family, BAX, BAK and BOK. The pore-forming

properties of BAX/BAK are controlled by a complex network of interactions with Bcl-2-family members, while those of BOK are mainly controlled through its rapid proteasomal degradation.

However, less well-known is the fact that Bcl-2-protein family members are able to regulate apoptosis through modulation of Ca^{2+}-signaling [226–228]. It is mainly the anti-apoptotic family members that partake in this moonlighting activity: Bcl-2 and Bcl-XL both interact with the IP_3R and modify its function. Bcl-2 inhibits IP_3R-mediated Ca^{2+}-release by binding to the central modulatory domain of the IP_3R through its BH4 domain and by suppressing IP_3R-mediated Ca^{2+}-signaling [229,230]. Additional interactions between the transmembrane domains of both Bcl-2 and the IP_3R [231] and between the ligand-binding domain of the IP_3R and the BH4 domain of Bcl-2 [232] contribute to the inhibition of IP_3R function. The functional implications of Bcl-XL interacting with IP_3Rs appears even more complex. Bcl-XL can stimulate mitochondrial bioenergetics by promoting oscillatory IP_3R-mediated Ca^{2+} signaling towards the mitochondria [18]. Bcl-XL through its hydrophobic cleft formed by its BH3-BH1-BH2 domains binds to the C-terminal channel pore region of IP_3Rs that has been proposed to possess 2 BH3-like domains [233]. Interference with the IP_3R/Bcl-XL complex either through BH3 mimetic drugs [233] or through proteins such as phosphorylated K-Ras [234] compromises mitochondrial bio-energetics and renders cells more susceptible to cell death. Bcl-XL can however also inhibit IP_3R-mediated Ca^{2+} signaling [235]. This discrepancy is not fully understood, though it has been reported that the sensitizing effect of Bcl-XL on IP_3Rs displays a bell-shaped dependent course with a maximal effect at 1 μM Bcl-XL [233].

A more recent addition to the modulators of ER-mitochondria Ca^{2+} signaling is BOK, a pro-apoptotic protein of the Bcl-2-protein family. BOK binds to IP_3R1 and IP_3R2, thereby protecting them from proteolytic degradation [236], while vice versa IP_3R-scaffolded BOK is stabilized and protected from proteasomal degradation [237,238]. Moreover, a recent study by the Katz lab has shown that BOK is necessary for the proper localization of the IP_3Rs to the MAMs, where BOK enhances ER-mitochondrial Ca^{2+} fluxes [239]. Hence, BOK can fulfil part of its pro-apoptotic role by boosting IP_3R-mediated ER-mitochondria Ca^{2+} transfer.

The interaction between Bcl-2-protein family members and the IP_3R becomes especially interesting in the setting of cancer. The former proteins are often overexpressed in cancer cells to elicit apoptotic resistance. It appears that at least part of this apoptotic resistance is conferred through Bcl-2's or Bcl-XL's effect on Ca^{2+} signaling. This is illustrated by the observation that peptide-mediated disruption of the Bcl-2-IP_3R interaction results in mitochondrial Ca^{2+} overload and cell death in a number of cancer cell models [240–243]. Additionally, Bcl-2-protein family members may play a role in chemoresistance in cancer cells through their Ca^{2+}-signaling-modulating properties, since many

clinically used chemotherapeutics affect Ca^{2+} signaling [244]. For example, cisplatin resistance in ovarian cancer cell lines can be overcome by suppressing the Bcl-2-dependent inhibition of IP_3R-mediated Ca^{2+} signaling [245].

A second layer of control over mitochondrial Ca^{2+} overload-induced apoptosis, is at the level of the Ca^{2+} uptake into the mitochondria. Like for IP_3Rs, expression levels, binding partners and post-translational modifications play a role in regulating mitochondrial Ca^{2+} uptake through VDAC1 and the MCU complex. VDAC1 forms also a target of anti-apoptotic Bcl-2-family members. Bcl-XL binds via its BH4 domain to VDAC1 and inhibits its channel activity, thereby limiting pro-apoptotic Ca^{2+} transfer into the mitochondria [246]. The most interesting developments have, however, been made at the level of the MCU. As discussed in section 1, the MCU is the channel pore that is part of a large protein complex located in the IMM. This complex normally consists of EMRE, MCUb, MCUR1, MICU1 and MICU2 [67,247,248]. In certain tissues, like the brain, MICU2 is replaced in the complex by its isoform MICU3 [249,250]. Both MCU and EMRE are necessary to form the functional Ca^{2+} channel in the IMM, while the other components of the complex regulate MCU's Ca^{2+} signaling properties. This regulation can have important consequences. For example, Lambert et al. show that during cardiac injury, the incorporation of dominant-negative MCUb in the MCU complex increases [251]. This is proposed to act as an adaptive mechanism to limit mitochondrial Ca^{2+} overload during ischemia/reperfusion injury. In the context of cancer, MCUb was also found to be a marker for bad prognosis for glioma. However, this appeared not to be related to remodeling of mitochondrial Ca^{2+} signaling [252]. MICU1, another protein of the MCU complex is overexpressed in ovarian cancer cell lines, leading to a similar effect as incorporation of MCUb [253]. Since MICU1 acts as a gatekeeper for MCU-mediated Ca^{2+} uptake, overexpression of MICU1 in cancer cells increases the threshold for mitochondrial Ca^{2+} uptake, and in this way elicits chemoresistance. In the same study, silencing of MICU1 in vivo was able to restore cisplatin sensitivity [253].

A last factor that determines the risk of mitochondrial Ca^{2+} overload is the distance between ER and mitochondria at the contact sites. The current paradigm prescribes that IP_3Rs at the ER and VDAC1 at the mitochondria are part of a protein complex at those contact sites (see section 3). This allows for the quasi-synaptic transfer of Ca^{2+} from the IP_3R to VDAC1 that shuttles Ca^{2+} to the IMS for further uptake through the MCU [44,254]. Since the MCU has a relatively low affinity for Ca^{2+} (K_d ~10 µM), the microdomain at the MAMs is crucial to locally concentrate Ca^{2+} to sufficiently high concentrations to allow uptake through the MCU. The extent to which such microdomains are present is therefore an important factor in cellular sensitivity to apoptosis. MAMs are considered to be highly dynamic, but little is known about how this dynamic nature is regulated at the molecular level [116]. In general, high levels of tethering proteins at the MAMs, which narrow the distance between ER and

mitochondria, increase the sensitivity towards mitochondrial Ca^{2+} overload [255,256]. On the contrary, depleting ER-mitochondria tethers has the opposite effect [85,257]. However, it is likely that dynamic responses of MAMs require more subtle control mechanisms than simply modulating protein tether levels. For example, local [Ca^{2+}] -and thus also Ca^{2+} release or Ca^{2+} leakage from the ER- may act as a regulator of ER-mitochondria contact sites [258,259]. What we do know, however, is that in pathological situations the distance between ER and mitochondria is often altered at the MAMs. For example, in cancer, the distance is often increased, protecting cancer cells from mitochondrial Ca^{2+} overload and demise [260,261]. This is one mechanism through which chemoresistance can occur. Alternatively, in AD, MAMs are thought to be tightened, increasing the sensitivity of neurons to mitochondrial Ca^{2+} overload and contributing to the neuronal cell death that is a hallmark of the disease [73,141].

Given the modulation of ER-mitochondria Ca^{2+}-transfer in pathological contexts, it is not surprising that scientists regard targeting mitochondrial Ca^{2+} uptake and its associated consequences as a viable option. A classic example in the field is ischemia/reperfusion injury, where it is shown that it is not necessarily the ischemic period that is detrimental for cells, but rather the reperfusion afterwards [262,263]. This reperfusion is accompanied by mitochondrial ROS production and mitochondrial Ca^{2+} overload, which eventually leads to cell death [264]. Numerous studies have shown that controlling mitochondrial Ca^{2+} uptake is able to limit the damage upon ischemia/reperfusion injury [265–267]. Earlier studies used cyclosporine A to target and inhibit the mPTP directly [268], whereas more recent studies have shown that blocking Ca^{2+} uptake via the MCU has a similar beneficial effect [265,269]. This is one of many examples through which insights in biological processes at the molecular level may pave the way towards new therapeutic approaches.

6. Conclusions

The ER-mitochondria contact sites are established and sustained by tethering proteins. By serving as hotspots for Ca^{2+} signaling microdomains between ER and mitochondria, they play a pivotal role in cell function and particularly in cell death and survival. ER-resident IP_3R channels are present in the MAMs, where they are linked via the chaperone GRP75 to the OMM-resident VDAC1. At the MAMs, IP_3Rs fulfil two functions: first, they serve as ER-mitochondria tethers contributing to the formation of ER-mitochondrial contact sites; second, they engage in ER-mitochondrial Ca^{2+} transfer, establishing a corridor for quasi-synaptic Ca^{2+} transfer. Although IP_3Rs can traffic in and out of the MAMs, when parked there, they become particularly effective in mediating ER-mitochondria Ca^{2+} transfer. As such, IP_3Rs critically control mitochondrial functions, from cell metabolism to cell death. During the last years, a large number of proteins have been identified to reside at contact sites and to influence contact site formation. Several of these proteins directly impact the Ca^{2+}-flux properties, the

tethering function and/or abundance of IP_3Rs at MAMs. These proteins ensure a tight control of Ca^{2+} delivery towards the mitochondria. Moreover, several of these accessory proteins are directly implicated in diseases, whereby loss-of-function or gain-of-function mutations underlies pathological conditions such as neurodegenerative diseases, metabolic diseases, muscle diseases and cancer. As a consequence, aberrations in the levels or in the localization of such MAM-resident proteins impact ER-mitochondria contact site properties (extent of contacts, distance between ER and mitochondria, number of contacts) and provoke dysregulation of Ca^{2+} signaling at the ER-mitochondria Ca^{2+} interface that translate into mitochondrial defects. The output can be compromised mitochondrial functionality and bio-energetics (as in the case of Wolfram syndrome or PD), cell death resistance (as in cancers) when ER-mitochondria Ca^{2+} transfer are severely hampered, or can be excessive cell death susceptibility (as in neurodegenerative diseases such as AD) when ER-mitochondria Ca^{2+} transfer is excessively boosted. This has important ramifications. First, dysregulation of MAM architecture and functionality is an important driver of the pathogenesis in a broad set of diseases at the subcellular and microdomain level. Second, restoring MAM properties can reset mitochondrial functionality, thereby preventing pathogenesis and/or dampen disease burden. Third, the rich network of proteins and/or accessory proteins contributing to MAM architecture and functionality also offer attractive, and potentially pharmacologically tangible, targets and exciting avenues to develop novel therapeutic strategies to fix MAM defects, to restore mitochondrial functionality and thus improve cellular health. For instance, activation of the MAM-resident S1R or elevating NCS-1-protein levels can restore the MAM functionality and ER-mitochondria Ca^{2+} fluxes in Wolfram syndrome, thereby alleviating cellular and systemic/behavioral deficits arising from loss of WFS1. Instead, in AD, which is characterized by a tightening of MAMs with subsequent mitochondrial Ca^{2+} overload, spacing ER from mitochondria alleviates Aβ42-driven neurotoxicity and improves behavioral outcomes. It will be exciting to further explore the role of MAMs in pathogenesis and their therapeutic potential using pharmacological and/or genetic means to tackle diseases.

Acknowledgements

Research in the laboratory of the authors was supported by Grants from the Research Foundation—Flanders (FWO) (G0C9114N, G0A3416N, G090118N, G0A6919N, G081821N, G094522N), by the Research Council of the KU Leuven (Grants OT14/101, C14/19/099 and AKUL/19/34). IdR is recipient of a Ph.D. fellowship from the FWO and MK is holder of a Marie Curie fellowship.

Legends to the figures

Fig. 1. Overview of the proteins involved in the tethering of the ER to the mitochondria. ER-localized IP_3Rs are coupled to VDAC1 in the outer mitochondrial membrane (OMM) via the chaperone protein GRP75 thereby forming a Ca^{2+}-transporting 'highway'. In cardiomyocytes, IP_3R2 interacts with FUNDC1 at the OMM, thus increasing ER-mitochondria tethering. Mitofusin (MFN) 2 is located at both the OMM and the ER and is able to form either a homodimer or a heterodimer with MFN1, which is solely located at the OMM, thereby forming a bridge across the MAMs. Another tethering protein complex that directly increase MAM formation and its size is formed by the ER protein VAPB and mitochondrial PTPIP51. BAP31 in the ER membrane interacts with the fission protein Fis1 in the OMM, whereby ER-localized PACS-2 regulates BAP31s MAM interaction. Finally, PDZD8 localized at the ER membrane is a known tethering protein that increases MAM formation. However, its mitochondrial tethering partner is yet to be identified. Proteins with white lettering function as Ca^{2+} channels. See text for more information. Created with BioRender.com.

Fig. 2. Overview of the complex consisting of the inositol 1,4,5-trisphosphate receptor (IP_3R) in the ER, glucose-regulated protein 75 (GRP75) and the voltage-dependent anion channel 1 (VDAC1) in the outer mitochondrial membrane (OMM) with the main interacting partners controlling ER-mitochondria tethering and/or Ca^{2+} flux. This complex forms together with the mitochondrial calcium uniporter (MCU) in the inner mitochondrial membrane (IMM) the Ca^{2+}-transporting axis. Parkinson's disease related protein DJ-1 interacts with all partners of the IP_3R3-GRP75-VDAC1 complex and regulates IP_3R3 degradation. OMM-localized transglutaminase 2 (TG2) interacts with VDAC1 and GRP75 thereby indirectly regulating the interaction between GRP75 and IP_3R3. TOM70 is located on the OMM and interacts with IP_3R3 in order to recruit it to the mitochondria-associated membranes (MAMs), thereby facilitating IP_3R3-GRP75-VDAC1 complex formation. Thymocyte-expressed positive selection-associated gene 1 (TESPA1) interacts with the N-terminal domain of IP_3R1 and IP_3R3, while also interacting with GRP75 and thus promoting MAM formation and stability. In the context of insulin resistance in skeletal muscle, PDK4 interacts with IP_3R1 and GRP75 to increase their tethering function. The presence of IRE1α and the activation of the sigma-1 receptor (S1R) in the ER membrane, increase MAM formation as well as the Ca^{2+} flux through the Ca^{2+}-transporting axis. S1R directly interacts with both the IP_3R and IRE1α, thereby stabilizing them. In turn, IRE1α is able to scaffold and stabilize IP_3R at the MAMs through a direct interaction via its C-terminal cytoplasmic region, independently of its enzymatic activity. It is possible that IRE1α, S1R, and IP_3R form a tripartite complex regulating the tethering of ER to mitochondria, but this remains to be proven. BCL-

2 is an anti-apoptotic protein suppressing Ca^{2+} flux through the IP_3R, preventing mitochondrial Ca^{2+} overload. Lastly, Wolfram syndrome 1 (WFS1) is believed to form a complex with the IP_3R through NCS-1. The interaction between WFS1 and NCS-1 prevents the degradation of the latter. Both WFS1 and NCS-1 enhance Ca^{2+} flux through the Ca^{2+}-transporting axis while WFS1 also supports both MAM formation and mitochondrial bioenergetics. Proteins with white lettering function as Ca^{2+} channels. See text for more information. Created with BioRender.com.

References

[1] A. Verkhratsky, Physiology and pathophysiology of the calcium store in the endoplasmic reticulum of neurons, Physiol. Rev. 85 (2005) 201–279. https://doi.org/10.1152/physrev.00004.2004.

[2] J. Chen, A. Sitsel, V. Benoy, M.R. Sepúlveda, P. Vangheluwe, Primary active Ca^{2+} transport systems in health and disease, Cold Spring Harb. Perspect. Biol. 12 (2020) a035113. https://doi.org/10.1101/cshperspect.a035113.

[3] A. Schulte, R. Blum, Shaped by leaky ER: Homeostatic Ca^{2+} fluxes, Front. Physiol. 13 (2022) 972104. https://doi.org/10.3389/fphys.2022.972104.

[4] M. Zajac, S. Modi, Y. Krishnan, The evolution of organellar calcium mapping technologies, Cell Calcium. 108 (2022) 102658. https://doi.org/10.1016/j.ceca.2022.102658.

[5] J.K. Foskett, C. White, K.H. Cheung, D.O.D. Mak, Inositol trisphosphate receptor Ca^{2+} release channels, Physiol. Rev. 87 (2007) 593–658. https://doi.org/10.1152/physrev.00035.2006.

[6] K. Hamada, K. Mikoshiba, IP_3 receptor plasticity underlying diverse functions, Annu. Rev. Physiol. 82 (2020) 151–176. https://doi.org/10.1146/annurev-physiol-021119-034433.

[7] K.A. Woll, F. Van Petegem, Calcium-release channels: Structure and function of IP_3 receptors and ryanodine receptors, Physiol. Rev. 102 (2022) 209–268. https://doi.org/10.1152/PHYSREV.00033.2020.

[8] G. Santulli, D. Lewis, A. des Georges, A.R. Marks, J. Frank, Ryanodine receptor structure and function in health and disease, Subcell. Biochem 87 (2018) 329–352. https://doi.org/10.1007/978-981-10-7757-9_11.

[9] D. Gong, N. Yan, H.A. Ledford, Structural basis for the modulation of ryanodine receptors, Trends Biochem. Sci. 46 (2021) 489–501. https://doi.org/10.1016/j.tibs.2020.11.009.

[10] F.O. Lemos, G. Bultynck, J.B. Parys, A comprehensive overview of the complex world of the endo- and sarcoplasmic reticulum Ca^{2+}-leak channels, Biochim. Biophys. Acta - Mol. Cell Res. 1868 (2021) 119020. https://doi.org/10.1016/j.bbamcr.2021.119020.

[11] K.J. Alzayady, L. Wang, R. Chandrasekhar, L.E. Wagner, F. Van Petegem, D.I. Yule, Defining the stoichiometry of inositol 1,4,5-trisphosphate binding required to initiate Ca^{2+} release, Sci. Signal. 9 (2016) ra35. https://doi.org/10.1126/SCISIGNAL.AAD6281.

[12] T. Miyakawa, A. Maeda, T. Yamazawa, K. Hirose, T. Kurosaki, M. Iino, Encoding of Ca^{2+} signals

by differential expression of IP_3 receptor subtypes, EMBO J. 18 (1999) 1303–1308. https://doi.org/10.1093/emboj/18.5.1303.

[13] V. Vanderheyden, B. Devogelaere, L. Missiaen, H. De Smedt, G. Bultynck, J.B. Parys, Regulation of inositol 1,4,5-trisphosphate-induced Ca^{2+} release by reversible phosphorylation and dephosphorylation, Biochim. Biophys. Acta. 1793 (2009) 959. https://doi.org/10.1016/J.BBAMCR.2008.12.003.

[14] D.L. Prole, C.W. Taylor, Inositol 1,4,5-trisphosphate receptors and their protein partners as signalling hubs, J. Physiol. 594 (2016) 2849–2866. https://doi.org/10.1113/JP271139.

[15] J.B. Parys, T. Vervliet, New insights in the IP_3 receptor and its regulation, Adv. Exp. Med. Biol. 1131 (2020) 243–270. https://doi.org/10.1007/978-3-030-12457-1_10.

[16] N.B. Thillaiappan, H.A. Smith, P. Atakpa-Adaji, C.W. Taylor, KRAP tethers IP_3 receptors to actin and licenses them to evoke cytosolic Ca^{2+} signals, Nat. Commun. 12 (2021) 4514. https://doi.org/10.1038/s41467-021-24739-9.

[17] Y.P. Rong, A.S. Aromolaran, G. Bultynck, F. Zhong, X. Li, K. McColl, S. Matsuyama, S. Herlitze, H.L. Roderick, M.D. Bootman, G.A. Mignery, J.B. Parys, H. De Smedt, C.W. Distelhorst, Targeting Bcl-2-IP_3 receptor interaction to reverse Bcl-2's inhibition of apoptotic calcium signals, Mol. Cell. 31 (2008) 255–265. https://doi.org/10.1016/j.molcel.2008.06.014.

[18] C. White, C. Li, J. Yang, N.B. Petrenko, M. Madesh, C.B. Thompson, J.K. Foskett, The endoplasmic reticulum gateway to apoptosis by Bcl-XL modulation of the $InsP_3R$, Nat. Cell Biol. 7 (2005) 1021–1028. https://doi.org/10.1038/ncb1302.

[19] A. Bononi, C. Giorgi, S. Patergnani, D. Larson, K. Verbruggen, M. Tanji, L. Pellegrini, V. Signorato, F. Olivetto, S. Pastorino, M. Nasu, A. Napolitano, G. Gaudino, P. Morris, G. Sakamoto, L.K. Ferris, A. Danese, A. Raimondi, C. Tacchetti, S. Kuchay, H.I. Pass, E.B. Affar, H. Yang, P. Pinton, M. Carbone, BAP1 regulates IP_3R3-mediated Ca^{2+} flux to mitochondria suppressing cell transformation, Nature. 546 (2017) 549–553. https://doi.org/10.1038/nature22798.

[20] S. Kuchay, C. Giorgi, D. Simoneschi, J. Pagan, S. Missiroli, A. Saraf, L. Florens, M.P. Washburn, A. Collazo-Lorduy, M. Castillo-Martin, C. Cordon-Cardo, S.M. Sebti, P. Pinton, M. Pagano, PTEN counteracts FBXL2 to promote IP_3R3- and Ca^{2+}-mediated apoptosis limiting tumour growth, Nature. 546 (2017) 554–558. https://doi.org/10.1038/nature22965.

[21] X. Gao, C.G. Bonzerato, R.J.H. Wojcikiewicz, Binding of the erlin1/2 complex to the third

intralumenal loop of IP_3R1 triggers its ubiquitin-proteasomal degradation, J. Biol. Chem. 298 (2022) 102026. https://doi.org/10.1016/j.jbc.2022.102026.

[22] Y. Xue, J.L. Morris, K. Yang, Z. Fu, X. Zhu, F. Johnson, B. Meehan, L. Witkowski, A. Yasmeen, T. Golenar, M. Coatham, G. Morin, A. Monast, V. Pilon, P.O. Fiset, S. Jung, A. V. Gonzalez, S. Camilleri-Broet, L. Fu, L.M. Postovit, J. Spicer, W.H. Gotlieb, M.C. Guiot, J. Rak, M. Park, W. Lockwood, W.D. Foulkes, J. Prudent, S. Huang, SMARCA4/2 loss inhibits chemotherapy-induced apoptosis by restricting IP_3R3-mediated Ca^{2+} flux to mitochondria, Nat. Commun. 12 (2021) 5404. https://doi.org/10.1038/s41467-021-25260-9.

[23] L.E. Wagner, S.K. Joseph, D.I. Yule, Regulation of single inositol 1,4,5-trisphosphate receptor channel activity by protein kinase A phosphorylation, J. Physiol. 586 (2008) 3577–3596. https://doi.org/10.1113/jphysiol.2008.152314.

[24] T. Szado, V. Vanderheyden, J.B. Parys, H. De Smedt, K. Rietdorf, L. Kotelevets, E. Chastre, F. Khan, U. Landegren, O. Söderberg, M.D. Bootman, H.L. Roderick, Phosphorylation of inositol 1,4,5-trisphosphate receptors by protein kinase B/Akt inhibits Ca^{2+} release and apoptosis, Proc. Natl. Acad. Sci. U. S. A. 105 (2008) 2427–2432. https://doi.org/10.1073/pnas.0711324105.

[25] S. Marchi, A. Rimessi, C. Giorgi, C. Baldini, L. Ferroni, R. Rizzuto, P. Pinton, Akt kinase reducing endoplasmic reticulum Ca^{2+} release protects cells from Ca^{2+}-dependent apoptotic stimuli, Biochem. Biophys. Res. Commun. 375 (2008) 501–505. https://doi.org/10.1016/j.bbrc.2008.07.153.

[26] M.D. Bootman, G. Bultynck, Fundamentals of cellular calcium signaling: A primer, Cold Spring Harb. Perspect. Biol. 12 (2020) a038802. https://doi.org/10.1101/cshperspect.a038802.

[27] J. Humeau, J.M. Bravo-San Pedro, I. Vitale, L. Nuñez, C. Villalobos, G. Kroemer, L. Senovilla, Calcium signaling and cell cycle: Progression or death, Cell Calcium. 70 (2018) 3–15. https://doi.org/10.1016/j.ceca.2017.07.006.

[28] S. Patergnani, A. Danese, E. Bouhamida, G. Aguiari, M. Previati, P. Pinton, C. Giorgi, Various aspects of calcium signaling in the regulation of apoptosis, autophagy, cell proliferation, and cancer, Int. J. Mol. Sci. 21 (2020) 1–27. https://doi.org/10.3390/ijms21218323.

[29] A. Duxfield, J. Munkley, M.D. Briggs, E.P. Dennis, CRELD2 is a novel modulator of calcium release and calcineurin-NFAT signalling during osteoclast differentiation, Sci. Rep. 12 (2022) 13884. https://doi.org/10.1038/s41598-022-17347-0.

[30] S. Sinha, Y. Elbaz-Alon, O. Avinoam, Ca^{2+} as a coordinator of skeletal muscle differentiation, fusion and contraction, FEBS J. 289 (2022) 6531–6542. https://doi.org/10.1111/febs.16552.

[31] R.A. Seidman, H. Khattab, J.J. Polanco, J.E. Broome, F.J. Sim, Oscillatory calcium release and sustained store-operated oscillatory calcium signaling prevents differentiation of human oligodendrocyte progenitor cells, Sci. Rep. 12 (2022) 6160. https://doi.org/10.1038/s41598-022-10095-1.

[32] N. Martin, D. Bernard, Calcium signaling and cellular senescence, Cell Calcium. 70 (2018) 16–23. https://doi.org/10.1016/j.ceca.2017.04.001.

[33] U. Ahumada-Castro, A. Puebla-Huerta, V. Cuevas-Espinoza, A. Lovy, J.C. Cardenas, Keeping zombies alive: The ER-mitochondria Ca^{2+} transfer in cellular senescence, Biochim. Biophys. Acta - Mol. Cell Res. 1868 (2021) 119099. https://doi.org/10.1016/j.bbamcr.2021.119099.

[34] D. V. Ziegler, D. Vindrieux, D. Goehrig, S. Jaber, G. Collin, A. Griveau, C. Wiel, N. Bendridi, S. Djebali, V. Farfariello, N. Prevarskaya, L. Payen, J. Marvel, S. Aubert, J.M. Flaman, J. Rieusset, N. Martin, D. Bernard, Calcium channel ITPR2 and mitochondria–ER contacts promote cellular senescence and aging, Nat. Commun. 12 (2021) 720. https://doi.org/10.1038/s41467-021-20993-z.

[35] V. Farfariello, D. V. Gordienko, L. Mesilmany, Y. Touil, E. Germain, I. Fliniaux, E. Desruelles, D. Gkika, M. Roudbaraki, G. Shapovalov, L. Noyer, M. Lebas, L. Allart, N. Zienthal-Gelus, O. Iamshanova, F. Bonardi, M. Figeac, W. Laine, J. Kluza, P. Marchetti, B. Quesnel, D. Metzger, D. Bernard, J.B. Parys, L. Lemonnier, N. Prevarskaya, TRPC3 shapes the ER-mitochondria Ca^{2+} transfer characterizing tumour-promoting senescence, Nat. Commun. 13 (2022) 956. https://doi.org/10.1038/s41467-022-28597-x.

[36] R.M.L. La Rovere, G. Roest, G. Bultynck, J.B. Parys, Intracellular Ca^{2+} signaling and Ca^{2+} microdomains in the control of cell survival, apoptosis and autophagy, Cell Calcium. 60 (2016) 74–87. https://doi.org/10.1016/j.ceca.2016.04.005.

[37] E. Kania, G. Roest, T. Vervliet, J.B. Parys, G. Bultynck, IP_3 receptor-mediated calcium signaling and its role in autophagy in cancer, Front. Oncol. 7 (2017) 140. https://doi.org/10.3389/fonc.2017.00140.

[38] M.D. Bootman, T. Chehab, G. Bultynck, J.B. Parys, K. Rietdorf, The regulation of autophagy by calcium signals: Do we have a consensus?, Cell Calcium. 70 (2018) 32–46. https://doi.org/10.1016/j.ceca.2017.08.005.

[39] P. Sukumaran, V.N. Da Conceicao, Y. Sun, N. Ahamad, L.R. Saraiva, S. Selvaraj, B.B. Singh, Calcium signaling regulates autophagy and apoptosis, Cells. 10 (2021) 2125. https://doi.org/10.3390/cells10082125.

[40] Q. Zheng, Y. Chen, D. Chen, H. Zhao, Y. Feng, Q. Meng, Y. Zhao, H. Zhang, Calcium transients on the ER surface trigger liquid-liquid phase separation of FIP200 to specify autophagosome initiation sites, Cell. 185 (2022) 4082-4098.e22. https://doi.org/10.1016/j.cell.2022.09.001.

[41] S.K. Joseph, D.M. Booth, M.P. Young, G. Hajnóczky, Redox regulation of ER and mitochondrial Ca^{2+} signaling in cell survival and death, Cell Calcium. 79 (2019) 89–97. https://doi.org/10.1016/j.ceca.2019.02.006.

[42] A. Danese, S. Leo, A. Rimessi, M.R. Wieckowski, F. Fiorica, C. Giorgi, P. Pinton, Cell death as a result of calcium signaling modulation: A cancer-centric prospective, Biochim. Biophys. Acta - Mol. Cell Res. 1868 (2021) 119061. https://doi.org/10.1016/j.bbamcr.2021.119061.

[43] G. Csordás, D. Weaver, G. Hajnóczky, Endoplasmic reticulum–mitochondrial contactology: Structure and signaling functions, Trends Cell Biol. 28 (2018) 523–540. https://doi.org/10.1016/j.tcb.2018.02.009.

[44] M.L. Sassano, B. Felipe-Abrio, P. Agostinis, ER-mitochondria contact sites; a multifaceted factory for Ca^{2+} signaling and lipid transport, Front. Cell Dev. Biol. 10 (2022) 988014. https://doi.org/10.3389/fcell.2022.988014.

[45] T. Hayashi, R. Rizzuto, G. Hajnoczky, T.P. Su, MAM: more than just a housekeeper, Trends Cell Biol. 19 (2009) 81–88. https://doi.org/10.1016/j.tcb.2008.12.002.

[46] J.R. Friedman, L.L. Lackner, M. West, J.R. DiBenedetto, J. Nunnari, G.K. Voeltz, ER tubules mark sites of mitochondrial division, Science. 334 (2011) 358–362. https://doi.org/10.1126/science.1207385.

[47] M. Hamasaki, N. Furuta, A. Matsuda, A. Nezu, A. Yamamoto, N. Fujita, H. Oomori, T. Noda, T. Haraguchi, Y. Hiraoka, A. Amano, T. Yoshimori, Autophagosomes form at ER-mitochondria contact sites, Nature. 495 (2013) 389–393. https://doi.org/10.1038/nature11910.

[48] A.R. van Vliet, T. Verfaillie, P. Agostinis, New functions of mitochondria associated membranes in cellular signaling, Biochim. Biophys. Acta - Mol. Cell Res. 1843 (2014) 2253–2262. https://doi.org/10.1016/j.bbamcr.2014.03.009.

[49] C. Ji, Z. Zhang, Z. Li, X. She, X. Wang, B. Li, X. Xu, D. Song, D. Zhang, Mitochondria-associated endoplasmic reticulum membranes: Inextricably linked with autophagy process, Oxid. Med.

Cell. Longev. 2022 (2022) 7086807. https://doi.org/10.1155/2022/7086807.

[50] S.T. Degechisa, Y.T. Dabi, S.T. Gizaw, The mitochondrial associated endoplasmic reticulum membranes: A platform for the pathogenesis of inflammation-mediated metabolic diseases, Immunity, Inflamm. Dis. 10 (2022) e647. https://doi.org/10.1002/iid3.647.

[51] G. Csordás, A.P. Thomas, G. Hajnóczky, Quasi-synaptic calcium signal transmission between endoplasmic reticulum and mitochondria, EMBO J. 18 (1999) 96–108. https://doi.org/10.1093/emboj/18.1.96.

[52] G. Szalait, G. Csordást, B.M. Hantash, A.P. Thomas, G. Hajnóczky, Calcium signal transmission between ryanodine receptors and mitochondria, J. Biol. Chem. 275 (2000) 15305–15313. https://doi.org/10.1074/jbc.275.20.15305.

[53] P. Pacher, A.P. Thomas, G. Hajnóczky, Ca^{2+} marks: miniature calcium signals in single mitochondria driven by ryanodine receptors, Proc. Natl. Acad. Sci. U. S. A. 99 (2002) 2380–2385. https://doi.org/10.1073/pnas.032423699.

[54] C. García-Pérez, G. Hajnóczky, G. Csordás, Physical coupling supports the local Ca^{2+} transfer between sarcoplasmic reticulum subdomains and the mitochondria in heart muscle, J. Biol. Chem. 283 (2008) 32771–32780. https://doi.org/10.1074/jbc.M803385200.

[55] C.K. Min, D.R. Yeom, K.E. Lee, H.K. Kwon, M. Kang, Y.S. Kim, Z.Y. Park, H. Jeon, D.H. Kim, Coupling of ryanodine receptor 2 and voltage-dependent anion channel 2 is essential for Ca^{2+} transfer from the sarcoplasmic reticulum to the mitochondria in the heart, Biochem. J. 447 (2012) 371–379. https://doi.org/10.1042/BJ20120705.

[56] M. Yi, D. Weaver, V. Eisner, P. Várnai, L. Hunyady, J. Ma, G. Csordás, G. Hajnóczky, Switch from ER-mitochondrial to SR-mitochondrial calcium coupling during muscle differentiation, Cell Calcium. 52 (2012) 355–365. https://doi.org/10.1016/j.ceca.2012.05.012.

[57] V. Eisner, G. Csordás, G. Hajnóczky, Interactions between sarco-endoplasmic reticulum and mitochondria in cardiac and skeletal muscle-pivotal roles in Ca^{2+} and reactive oxygen species signaling, J. Cell Sci. 126 (2013) 2965–2978. https://doi.org/10.1242/jcs.093609.

[58] M. Ruiz-Meana, M. Minguet, D. Bou-Teen, E. Miro-Casas, C. Castans, J. Castellano, E. Bonzon-Kulichenko, A. Igual, R. Rodriguez-Lecoq, J. Vázquez, D. Garcia-Dorado, Ryanodine receptor glycation favors mitochondrial damage in the senescent heart, Circulation. 139 (2019) 949–964. https://doi.org/10.1161/CIRCULATIONAHA.118.035869.

[59] N. Zanou, H. Dridi, S. Reiken, T. Imamura de Lima, C. Donnelly, U. De Marchi, M. Ferrini, J.

Vidal, L. Sittenfeld, J.N. Feige, P.M. Garcia-Roves, I.C. Lopez-Mejia, A.R. Marks, J. Auwerx, B. Kayser, N. Place, Acute RyR1 Ca^{2+} leak enhances NADH-linked mitochondrial respiratory capacity, Nat. Commun. 12 (2021) 7219. https://doi.org/10.1038/s41467-021-27422-1.

[60] D.M. Booth, B. Enyedi, M. Geiszt, P. Várnai, G. Hajnóczky, Redox nanodomains are induced by and control calcium signaling at the ER-mitochondrial interface, Mol. Cell. 63 (2016) 240–248. https://doi.org/10.1016/j.molcel.2016.05.040.

[61] M. Katona, Á. Bartók, Z. Nichtova, G. Csordás, E. Berezhnaya, D. Weaver, A. Ghosh, P. Várnai, D.I. Yule, G. Hajnóczky, Capture at the ER-mitochondrial contacts licenses IP_3 receptors to stimulate local Ca^{2+} transfer and oxidative metabolism, Nat. Commun. 13 (2022) 6779. https://doi.org/10.1038/s41467-022-34365-8.

[62] J.F. Garbincius, J.W. Elrod, Mitochondrial calcium exchange in physiology and disease , Physiol. Rev. 102 (2022) 893–992. https://doi.org/10.1152/physrev.00041.2020.

[63] V. Shoshan-Barmatz, Y. Krelin, A. Shteinfer-Kuzmine, VDAC1 functions in Ca^{2+} homeostasis and cell life and death in health and disease, Cell Calcium. 69 (2018) 81–100. https://doi.org/10.1016/j.ceca.2017.06.007.

[64] W.M. Rosencrans, M. Rajendran, S.M. Bezrukov, T.K. Rostovtseva, VDAC regulation of mitochondrial calcium flux: From channel biophysics to disease, Cell Calcium. 94 (2021) 102356. https://doi.org/10.1016/j.ceca.2021.102356.

[65] G. Csordás, T. Golenár, E.L. Seifert, K.J. Kamer, Y. Sancak, F. Perocchi, C. Moffat, D. Weaver, S. De la Fuente, R. Bogorad, V. Koteliansky, J. Adijanto, V.K. Mootha, G. Hajnóczky, MICU1 controls both the threshold and cooperative activation of the mitochondrial Ca^{2+} uniporter, Cell Metab. 17 (2013) 976–987. https://doi.org/10.1016/j.cmet.2013.04.020.

[66] M. Paillard, G. Csordás, G. Szanda, T. Golenár, V. Debattisti, A. Bartok, N. Wang, C. Moffat, E.L. Seifert, A. Spät, G. Hajnóczky, Tissue-specific mitochondrial decoding of cytoplasmic Ca^{2+} signals is controlled by the stoichiometry of MICU1/2 and MCU, Cell Rep. 18 (2017) 2291–2300. https://doi.org/10.1016/j.celrep.2017.02.032.

[67] B.R. Alevriadou, A. Patel, M. Noble, S. Ghosh, V.M. Gohil, P.B. Stathopulos, M. Madesh, Molecular nature and physiological role of the mitochondrial calcium uniporter channel, Am. J. Physiol. - Cell Physiol. 320 (2021) C465–C482. https://doi.org/10.1152/ajpcell.00502.2020.

[68] W. Wu, J. Zheng, Z. Jia, Structural characterization of the mitochondrial Ca^{2+} uniporter provides insights into Ca^{2+} uptake and regulation, IScience. 24 (2021) 914–927.

https://doi.org/10.1016/j.isci.2021.102895.

[69] J. Yoo, Structural basis of Ca^{2+} uptake by mitochondrial calcium uniporter in mitochondria: a brief review, BMB Rep. 55 (2022) 528–534. https://doi.org/10.5483/BMBRep.2022.55.11.134.

[70] A. De Mario, D. D'Angelo, G. Zanotti, A. Raffaello, C. Mammucari, The mitochondrial calcium uniporter (MCU) complex - A play in five acts., Cell Calcium. 112 (2023) 102720.

[71] R. Rizzuto, M. Brini, M. Murgia, T. Pozzan, Microdomains with high Ca^{2+} close to IP_3-sensitive channels that are sensed by neighboring mitochondria, Science. 262 (1993) 744–747. https://doi.org/10.1126/science.8235595.

[72] R. Rizzuto, P. Pinton, W. Carrington, F.S. Fay, K.E. Fogarty, L.M. Lifshitz, R.A. Tuft, T. Pozzan, Close contacts with the endoplasmic reticulum as determinants of mitochondrial Ca^{2+} responses, Science. 280 (1998) 1763–1766. https://doi.org/10.1126/science.280.5370.1763.

[73] J. Loncke, A. Kaasik, I. Bezprozvanny, J.B. Parys, M. Kerkhofs, G. Bultynck, Balancing ER-mitochondrial Ca^{2+} fluxes in health and disease, Trends Cell Biol. 31 (2021) 598–612. https://doi.org/10.1016/j.tcb.2021.02.003.

[74] Y.E. Li, J.R. Sowers, C. Hetz, J. Ren, Cell death regulation by MAMs: from molecular mechanisms to therapeutic implications in cardiovascular diseases, Cell Death Dis. 13 (2022) 504. https://doi.org/10.1038/s41419-022-04942-2.

[75] P. Morgado-Cáceres, G. Liabeuf, X. Calle, L. Briones, J.A. Riquelme, R. Bravo-Sagua, V. Parra, The aging of ER-mitochondria communication: A journey from undifferentiated to aged cells, Front. Cell Dev. Biol. 10 (2022) 946678. https://doi.org/10.3389/fcell.2022.946678.

[76] A. Carreras-Sureda, G. Kroemer, J.C. Cardenas, C. Hetz, Balancing energy and protein homeostasis at ER-mitochondria contact sites, Sci. Signal. 15 (2022) eabm7524. https://doi.org/10.1126/scisignal.abm7524.

[77] Y. Zhang, Y. Wu, M. Zhang, Z. Li, B. Liu, H. Liu, J. Hao, X. Li, Synergistic mechanism between the endoplasmic reticulum and mitochondria and their crosstalk with other organelles, Cell Death Discov. 9 (2023) 51. https://doi.org/10.1038/s41420-023-01353-w.

[78] G. Csordás, C. Renken, P. Várnai, L. Walter, D. Weaver, K.F. Buttle, T. Balla, C.A. Mannella, G. Hajnóczky, Structural and functional features and significance of the physical linkage between ER and mitochondria, J. Cell Biol. 174 (2006) 915–921. https://doi.org/10.1083/jcb.200604016.

[79] G. Csordás, P. Várnai, T. Golenár, S. Roy, G. Purkins, T.G. Schneider, T. Balla, G. Hajnóczky, Imaging interorganelle contacts and local calcium dynamics at the ER-mitochondrial interface, Mol. Cell. 39 (2010) 121–132. https://doi.org/10.1016/j.molcel.2010.06.029.

[80] M. Giacomello, I. Drago, M. Bortolozzi, M. Scorzeto, A. Gianelle, P. Pizzo, T. Pozzan, Ca^{2+} hot spots on the mitochondrial surface are generated by Ca^{2+} mobilization from stores, but not by Aactivation of store-operated Ca^{2+} channels, Mol. Cell. 38 (2010) 280–290. https://doi.org/10.1016/j.molcel.2010.04.003.

[81] Y. Fan, T. Simmen, Mechanistic connections between endoplasmic reticulum (ER) redox control and mitochondrial metabolism, Cells. 8 (2019) 1071. https://doi.org/10.3390/cells8091071.

[82] T. Simmen, J.E. Aslan, A.D. Blagoveshchenskaya, L. Thomas, L. Wan, Y. Xiang, S.F. Feliciangeli, C.H. Hung, C.M. Crump, G. Thomas, PACS-2 controls endoplasmic reticulum-mitochondria communication and Bid-mediated apoptosis, EMBO J. 24 (2005) 717–729. https://doi.org/10.1038/sj.emboj.7600559.

[83] R. Iwasawa, A.L. Mahul-Mellier, C. Datler, E. Pazarentzos, S. Grimm, Fis1 and Bap31 bridge the mitochondria-ER interface to establish a platform for apoptosis induction, EMBO J. 30 (2011) 556–568. https://doi.org/10.1038/emboj.2010.346.

[84] B. Wang, M. Nguyen, N.C. Chang, G.C. Shore, Fis1, Bap31 and the kiss of death between mitochondria and endoplasmic reticulum, EMBO J. 30 (2011) 451–452. https://doi.org/10.1038/emboj.2010.352.

[85] G. Szabadkai, K. Bianchi, P. Várnai, D. De Stefani, M.R. Wieckowski, D. Cavagna, A.I. Nagy, T. Balla, R. Rizzuto, Chaperone-mediated coupling of endoplasmic reticulum and mitochondrial Ca^{2+} channels, J. Cell Biol. 175 (2006) 901–911. https://doi.org/10.1083/jcb.200608073.

[86] A. Bartok, D. Weaver, T. Golenár, Z. Nichtova, M. Katona, S. Bánsághi, K.J. Alzayady, V.K. Thomas, H. Ando, K. Mikoshiba, S.K. Joseph, D.I. Yule, G. Csordás, G. Hajnóczky, IP_3 receptor isoforms differently regulate ER-mitochondrial contacts and local calcium transfer, Nat. Commun. 10 (2019). https://doi.org/10.1038/s41467-019-11646-3.

[87] R. Filadi, N.S. Leal, B. Schreiner, A. Rossi, G. Dentoni, C.M. Pinho, B. Wiehager, D. Cieri, T. Calì, P. Pizzo, M. Ankarcrona, TOM70 sustains cell bioenergetics by promoting IP_3R3-mediated ER to mitochondria Ca^{2+} transfer, Curr. Biol. 28 (2018) 369-382.e6. https://doi.org/10.1016/j.cub.2017.12.047.

[88] A. Carreras-Sureda, F. Jaña, H. Urra, S. Durand, D.E. Mortenson, A. Sagredo, G. Bustos, Y. Hazari, E. Ramos-Fernández, M.L. Sassano, P. Pihán, A.R. van Vliet, M. González-Quiroz, A.K. Torres, C. Tapia-Rojas, M. Kerkhofs, R. Vicente, R.J. Kaufman, N.C. Inestrosa, C. Gonzalez-Billault, R.L. Wiseman, P. Agostinis, G. Bultynck, F.A. Court, G. Kroemer, J.C. Cárdenas, C. Hetz, Non-canonical function of IRE1α determines mitochondria-associated endoplasmic reticulum composition to control calcium transfer and bioenergetics, Nat. Cell Biol. 21 (2019) 755–767. https://doi.org/10.1038/s41556-019-0329-y.

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

[90] T. Hayashi, T.P. Su, Sigma-1 receptor chaperones at the ER-mitochondrion interface regulate Ca^{2+} signaling and cell survival, Cell. 131 (2007) 596–610. https://doi.org/10.1016/j.cell.2007.08.036.

[91] T. Verfaillie, N. Rubio, A.D. Garg, G. Bultynck, R. Rizzuto, J.P. Decuypere, J. Piette, C. Linehan, S. Gupta, A. Samali, P. Agostinis, PERK is required at the ER-mitochondrial contact sites to convey apoptosis after ROS-based ER stress, Cell Death Differ. 19 (2012) 1880–1891. https://doi.org/10.1038/cdd.2012.74.

[92] M.L. Sassano, A.R. van Vliet, E. Vervoort, S. Van Eygen, C. Van den Haute, B. Pavie, J. Roels, J. V Swinnen, M. Spinazzi, L. Moens, K. Casteels, I. Meyts, P. Pinton, S. Marchi, L. Rochin, F. Giordano, B. Felipe-Abrio, P. Agostinis, PERK recruits E-Syt1 at ER-mitochondria contacts for mitochondrial lipid transport and respiration, J. Cell Biol. 222 (2023) e202206008. https://doi.org/10.1083/jcb.202206008.

[93] S. Wu, Q. Lu, Q. Wang, Y. Ding, Z. Ma, X. Mao, K. Huang, Z. Xie, M.H. Zou, Binding of FUN14 domain containing 1 with inositol 1,4,5-trisphosphate receptor in mitochondria-associated endoplasmic reticulum membranes maintains mitochondrial dynamics and function in hearts in vivo, Circulation. 136 (2017) 2248–2266. https://doi.org/10.1161/CIRCULATIONAHA.117.030235.

[94] D. Naon, M. Zaninello, M. Giacomello, T. Varanita, F. Grespi, S. Lakshminaranayan, A. Serafini, M. Semenzato, S. Herkenne, M.I. Hernández-Alvarez, A. Zorzano, D. De Stefani, G.W. Dorn, L. Scorrano, Critical reappraisal confirms that Mitofusin 2 is an endoplasmic reticulum-mitochondria tether, Proc. Natl. Acad. Sci. U. S. A. 113 (2016) 11249–11254. https://doi.org/10.1073/pnas.1606786113.

[95] O.M. De Brito, L. Scorrano, Mitofusin 2 tethers endoplasmic reticulum to mitochondria, Nature. 456 (2008) 605–610. https://doi.org/10.1038/nature07534.

[96] R. Filadi, E. Greotti, G. Turacchio, A. Luini, T. Pozzan, P. Pizzo, Mitofusin 2 ablation increases endoplasmic reticulum-mitochondria coupling, Proc. Natl. Acad. Sci. U. S. A. 112 (2015) E2174–E2181. https://doi.org/10.1073/pnas.1504880112.

[97] N.S. Leal, B. Schreiner, C.M. Pinho, R. Filadi, B. Wiehager, H. Karlström, P. Pizzo, M. Ankarcrona, Mitofusin-2 knockdown increases ER–mitochondria contact and decreases amyloid β-peptide production, J. Cell. Mol. Med. 20 (2016) 1686–1695. https://doi.org/10.1111/jcmm.12863.

[98] P.T.C. Wang, P.O. Garcin, M. Fu, M. Masoudi, P. St-Pierre, N. Panté, I.R. Nabi, Distinct mechanisms controlling rough and smooth endoplasmic reticulum contacts with mitochondria, J. Cell Sci. 128 (2015) 2759–2765. https://doi.org/10.1242/jcs.171132.

[99] L. Li, G. Gao, J. Shankar, B. Joshi, L.J. Foster, I.R. Nabi, P38 MAP kinase-dependent phosphorylation of the Gp78 E3 ubiquitin ligase controls ER-mitochondria association and mitochondria motility, Mol. Biol. Cell. 26 (2015) 3828–3840. https://doi.org/10.1091/mbc.E15-02-0120.

[100] A. Rossi, P. Pizzo, R. Filadi, Calcium, mitochondria and cell metabolism: A functional triangle in bioenergetics, Biochim. Biophys. Acta - Mol. Cell Res. 1866 (2019) 1068–1078. https://doi.org/10.1016/j.bbamcr.2018.10.016.

[101] R. Filadi, E. Greotti, G. Turacchio, A. Luini, T. Pozzan, P. Pizzo, Presenilin 2 modulates endoplasmic reticulum-mitochondria coupling by tuning the antagonistic effect of mitofusin 2, Cell Rep. 15 (2016) 2226–2238. https://doi.org/10.1016/j.celrep.2016.05.013.

[102] P. Cosson, A. Marchetti, M. Ravazzola, L. Orci, Mitofusin-2 independent juxtaposition of endoplasmic reticulum and mitochondria: an ultrastructural study, PLoS One. 7 (2012) e46293. https://doi.org/10.1371/journal.pone.0046293.

[103] R. Filadi, Di. Pendin, P. Pizzo, Mitofusin 2: From functions to disease, Cell Death Dis. 9 (2018) 330. https://doi.org/10.1038/s41419-017-0023-6.

[104] A. Sugiura, S. Nagashima, T. Tokuyama, T. Amo, Y. Matsuki, S. Ishido, Y. Kudo, H.M. McBride, T. Fukuda, N. Matsushita, R. Inatome, S. Yanagi, MITOL regulates endoplasmic reticulum-mitochondria contacts via Mitofusin2, Mol. Cell. 51 (2013) 20–34. https://doi.org/10.1016/j.molcel.2013.04.023.

[105] A. Tanaka, M.M. Cleland, S. Xu, D.P. Narendra, D.F. Suen, M. Karbowski, R.J. Youle, Proteasome and p97 mediate mitophagy and degradation of mitofusins induced by Parkin, J. Cell Biol. 191 (2010) 1367–1380. https://doi.org/10.1083/jcb.201007013.

[106] T. Calì, D. Ottolini, A. Negro, M. Brini, Enhanced parkin levels favor ER-mitochondria crosstalk and guarantee Ca^{2+} transfer to sustain cell bioenergetics, Biochim. Biophys. Acta - Mol. Basis Dis. 1832 (2013) 495–508. https://doi.org/10.1016/j.bbadis.2013.01.004.

[107] V. Basso, E. Marchesan, C. Peggion, J. Chakraborty, S. von Stockum, M. Giacomello, D. Ottolini, V. Debattisti, F. Caicci, E. Tasca, V. Pegoraro, C. Angelini, A. Antonini, A. Bertoli, M. Brini, E. Ziviani, Regulation of ER-mitochondria contacts by Parkin via Mfn2, Pharmacol. Res. 138 (2018) 43–56. https://doi.org/10.1016/j.phrs.2018.09.006.

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

[109] R. Stoica, K.J. De Vos, S. Paillusson, S. Mueller, R.M. Sancho, K.F. Lau, G. Vizcay-Barrena, W.L. Lin, Y.F. Xu, J. Lewis, D.W. Dickson, L. Petrucelli, J.C. Mitchell, C.E. Shaw, C.C.J. Miller, ER-mitochondria associations are regulated by the VAPB-PTPIP51 interaction and are disrupted by ALS/FTD-associated TDP-43, Nat. Commun. 5 (2014) 3996. https://doi.org/10.1038/ncomms4996.

[110] P. Gómez-Suaga, B.G. Pérez-Nievas, E.B. Glennon, D.H.W. Lau, S. Paillusson, G.M. Mórotz, T. Calì, P. Pizzo, W. Noble, C.C.J. Miller, The VAPB-PTPIP51 endoplasmic reticulum-mitochondria tethering proteins are present in neuronal synapses and regulate synaptic activity, Acta Neuropathol. Commun. 7 (2019) 35. https://doi.org/10.1186/s40478-019-0688-4.

[111] S.M. Martín-Guerrero, A. Markovinovic, G.M. Mórotz, S. Salam, W. Noble, C.C.J. Miller, Targeting ER-mitochondria signaling as a therapeutic target for frontotemporal dementia and related amyotrophic lateral sclerosis, Front. Cell Dev. Biol. 10 (2022) 915931. https://doi.org/10.3389/fcell.2022.915931.

[112] Y. Hirabayashi, S.K. Kwon, H. Paek, W.M. Pernice, M.A. Paul, J. Lee, P. Erfani, A. Raczkowski, D.S. Petrey, L.A. Pon, F. Polleux, ER-mitochondria tethering by PDZD8 regulates Ca^{2+} dynamics in mammalian neurons, Science. 358 (2017) 623–630. https://doi.org/10.1126/science.aan6009.

[113] Y. Elbaz-Alon, Y. Guo, N. Segev, M. Harel, D.E. Quinnell, T. Geiger, O. Avinoam, D. Li, J.

Nunnari, PDZD8 interacts with Protrudin and Rab7 at ER-late endosome membrane contact sites associated with mitochondria, Nat. Commun. 11 (2020) 3645. https://doi.org/10.1038/s41467-020-17451-7.

[114] M. Doghman-Bouguerra, V. Granatiero, S. Sbiera, I. Sbiera, S. Lacas-Gervais, F. Brau, M. Fassnacht, R. Rizzuto, E. Lalli, FATE 1 antagonizes calcium- and drug-induced apoptosis by uncoupling ER and mitochondria, EMBO Rep. 17 (2016) 1264–1280. https://doi.org/10.15252/embr.201541504.

[115] A. Beaulant, M. Dia, B. Pillot, M.A. Chauvin, J. Ji-cao, C. Durand, N. Bendridi, S. Chanon, A. Vieille-Marchiset, C.C. Da Silva, S. Patouraux, R. Anty, A. Iannelli, A. Tran, P. Gual, H. Vidal, L. Gomez, M. Paillard, J. Rieusset, Endoplasmic reticulum-mitochondria miscommunication is an early and causal trigger of hepatic insulin resistance and steatosis, J. Hepatol. 77 (2022) 710–722. https://doi.org/10.1016/j.jhep.2022.03.017.

[116] M. Pichla, F. Sneyers, K.B. Stopa, G. Bultynck, M. Kerkhofs, Dynamic control of mitochondria-associated membranes by kinases and phosphatases in health and disease, Cell. Mol. Life Sci. 78 (2021) 6541–6556. https://doi.org/10.1007/s00018-021-03920-9.

[117] M. D'Eletto, F. Rossin, L. Occhigrossi, M.G. Farrace, D. Faccenda, R. Desai, S. Marchi, G. Refolo, L. Falasca, M. Antonioli, F. Ciccosanti, G.M. Fimia, P. Pinton, M. Campanella, M. Piacentini, Transglutaminase type 2 regulates ER-mitochondria contact sites by interacting with GRP75, Cell Rep. 25 (2018) 3573-3581.e4. https://doi.org/10.1016/j.celrep.2018.11.094.

[118] R. Wadhwa, K. Taira, S.C. Kaul, An Hsp70 family chaperone, mortalin/mthsp70/PBP74/Grp75: What, when, and where?, Cell Stress Chaperones. 7 (2002) 309–316. https://doi.org/10.1379/1466-1268(2002)007<0309:AHFCMM>2.0.CO;2.

[119] R. Rizzuto, S. Marchi, M. Bonora, P. Aguiari, A. Bononi, D. De Stefani, C. Giorgi, S. Leo, A. Rimessi, R. Siviero, E. Zecchini, P. Pinton, Ca^{2+} transfer from the ER to mitochondria: When, how and why, Biochim. Biophys. Acta - Bioenerg. 1787 (2009) 1342–1351. https://doi.org/10.1016/j.bbabio.2009.03.015.

[120] H. Xu, N. Guan, Y.L. Ren, Q.J. Wei, Y.H. Tao, G.S. Yang, X.Y. Liu, D.F. Bu, Y. Zhang, S.N. Zhu, IP_3R-Grp75-VDAC1-MCU calcium regulation axis antagonists protect podocytes from apoptosis and decrease proteinuria in an Adriamycin nephropathy rat model, BMC Nephrol. 19 (2018) 140. https://doi.org/10.1186/s12882-018-0940-3.

[121] B. Honrath, I. Metz, N. Bendridi, J. Rieusset, C. Culmsee, A.M. Dolga, Glucose-regulated

protein 75 determines ER–mitochondrial coupling and sensitivity to oxidative stress in neuronal cells, Cell Death Discov. 3 (2017) 17076. https://doi.org/10.1038/cddiscovery.2017.76.

[122] T. Liang, W. Hang, J. Chen, Y. Wu, B. Wen, K. Xu, B. Ding, J. Chen, ApoE4 (Δ272–299) induces mitochondrial-associated membrane formation and mitochondrial impairment by enhancing GRP75-modulated mitochondrial calcium overload in neuron, Cell Biosci. 11 (2021) 50. https://doi.org/10.1186/s13578-021-00563-y.

[123] H. Lin, Y. Peng, J. Li, Z. Wang, S. Chen, X. Qing, F. Pu, M. Lei, Z. Shao, Reactive oxygen species regulate endoplasmic reticulum stress and ER-mitochondrial Ca^{2+} crosstalk to promote programmed necrosis of rat nucleus pulposus cells under compression, Oxid. Med. Cell. Longev. (2021) 8810698. https://doi.org/10.1155/2021/8810698.

[124] T. Wang, Q. Zhu, B. Cao, Y. Cai, S. Wen, J. Bian, H. Zou, R. Song, J. Gu, X. Liu, Z. Liu, Y. Yuan, Ca^{2+} transfer via the ER-mitochondria tethering complex in neuronal cells contribute to cadmium-induced autophagy, Cell Biol. Toxicol. 38 (2022) 469–485. https://doi.org/10.1007/s10565-021-09623-y.

[125] Y. Liu, X. Ma, H. Fujioka, J. Liu, S. Chen, X. Zhu, DJ-1 regulates the integrity and function of ER-mitochondria association through interaction with IP3R3-Grp75-VDAC1, Proc. Natl. Acad. Sci. U. S. A. 116 (2019) 25322–25328. https://doi.org/10.1073/PNAS.1906565116.

[126] V. Basso, E. Marchesan, E. Ziviani, A trio has turned into a quartet: DJ-1 interacts with the IP_3R-Grp75-VDAC complex to control ER-mitochondria interaction, Cell Calcium. 87 (2020) 102186. https://doi.org/10.1016/J.CECA.2020.102186.

[127] I. Tai-Nagara, S. Matsuoka, H. Ariga, T. Suda, Mortalin and DJ-1 coordinately regulate hematopoietic stem cell function through the control of oxidative stress, Blood. 123 (2014) 41–50. https://doi.org/10.1182/blood-2013-06-508333.

[128] K. Hamada, A. Terauchi, K. Nakamura, T. Higo, N. Nukina, N. Matsumoto, C. Hisatsune, T. Nakamura, K. Mikoshiba, Aberrant calcium signaling by transglutaminase-mediated posttranslational modification of inositol 1,4,5-trisphosphate receptors, Proc. Natl. Acad. Sci. U. S. A. 111 (2014) E3966–E3975. https://doi.org/10.1073/pnas.1409730111.

[129] A.G. Erustes, M. D'Eletto, G.C. Guarache, R.P. Ureshino, C. Bincoletto, G.J. da Silva Pereira, M. Piacentini, S.S. Smaili, Overexpression of α-synuclein inhibits mitochondrial Ca^{2+} trafficking between the endoplasmic reticulum and mitochondria through MAMs by altering the GRP75–

IP_3R interaction, J. Neurosci. Res. 99 (2021) 2932–2947. https://doi.org/10.1002/jnr.24952.

[130] C. Angebault, J. Fauconnier, S. Patergnani, J. Rieusset, A. Danese, C.A. Affortit, J. Jagodzinska, C. Mégy, M. Quiles, C. Cazevieille, J. Korchagina, D. Bonnet-Wersinger, D. Milea, C. Hamel, P. Pinton, M. Thiry, A. Lacampagne, B. Delprat, C. Delettre, ER-mitochondria cross-talk is regulated by the Ca^{2+} sensor NCS1 and is impaired in Wolfram syndrome, Sci. Signal. 11 (2018) eaaq1380. https://doi.org/10.1126/scisignal.aaq1380.

[131] J. Loncke, T. Vervliet, J.B. Parys, A. Kaasik, G. Bultynck, Uniting the divergent Wolfram syndrome–linked proteins WFS1 and CISD2 as modulators of Ca^{2+} signaling, Sci. Signal. 14 (2021) 6165. https://doi.org/10.1126/scisignal.abc6165.

[132] C. Schlecker, W. Boehmerle, A. Jeromin, B. DeGray, A. Varshney, Y. Sharma, K. Szigeti-Buck, B.E. Ehrlich, Neuronal calcium sensor-1 enhancement of $InsP_3$ receptor activity is inhibited by therapeutic levels of lithium, J. Clin. Invest. 116 (2006) 1668–1674. https://doi.org/10.1172/JCI22466.

[133] W. Boehmerle, K. Zhang, M. Sivula, F.M. Heidrich, Y. Lee, S.E. Jordt, B.E. Ehrlich, Chronic exposure to paclitaxel diminishes phosphoinositide signaling by calpain-mediated neuronal calcium sensor-1 degradation, Proc. Natl. Acad. Sci. U. S. A. 104 (2007) 11103–11108. https://doi.org/10.1073/pnas.0701546104.

[134] L. Crouzier, E.M. Richard, C. Diez, M. Denus, A. Peyrel, H. Alzaeem, N. Cubedo, T. Delaunay, T. Maurice, B. Delprat, NCS1 overexpression restored mitochondrial activity and behavioral alterations in a zebrafish model of Wolfram syndrome, Mol. Ther. - Methods Clin. Dev. 27 (2022) 295–308. https://doi.org/10.1016/j.omtm.2022.10.003.

[135] L. Crouzier, A. Danese, Y. Yasui, E.M. Richard, J.C. Liévens, S. Patergnani, S. Couly, C. Diez, M. Denus, N. Cubedo, M. Rossel, M. Thiry, T.P. Su, P. Pinton, T. Maurice, B. Delprat, Activation of the sigma-1 receptor chaperone alleviates symptoms of Wolfram syndrome in preclinical models, Sci. Transl. Med. 14 (2022) eabh3763. https://doi.org/10.1126/scitranslmed.abh3763.

[136] E. Area-Gomez, M. Del Carmen Lara Castillo, M.D. Tambini, C. Guardia-Laguarta, A.J.C. De Groof, M. Madra, J. Ikenouchi, M. Umeda, T.D. Bird, S.L. Sturley, E.A. Schon, Upregulated function of mitochondria-associated ER membranes in Alzheimer disease, EMBO J. 31 (2012) 4106–4123. https://doi.org/10.1038/emboj.2012.202.

[137] L. Hedskog, C.M. Pinho, R. Filadi, A. Rönnbäck, L. Hertwig, B. Wiehager, P. Larssen, S. Gellhaar, A. Sandebring, M. Westerlund, C. Graff, B. Winblad, D. Galter, H. Behbahani, P. Pizzo, E.

Glaser, M. Ankarcrona, Modulation of the endoplasmic reticulum-mitochondria interface in Alzheimer's disease and related models, Proc. Natl. Acad. Sci. U. S. A. 110 (2013) 7916–7921. https://doi.org/10.1073/pnas.1300677110.

[138] T. Fernandes, R. Resende, D.F. Silva, A.P. Marques, A.E. Santos, S.M. Cardoso, M. Rosário Domingues, P.I. Moreira, C.F. Pereira, Structural and functional alterations in mitochondria-associated membranes (MAMs) and in mitochondria activate stress response mechanisms in an in vitro model of alzheimer's disease, Biomedicines. 9 (2021) 108827. https://doi.org/10.3390/biomedicines9080881.

[139] M. Calvo-Rodriguez, E.K. Kharitonova, B.J. Bacskai, In vivo brain imaging of mitochondrial Ca^{2+} in neurodegenerative diseases with multiphoton microscopy, Biochim. Biophys. Acta - Mol. Cell Res. 1868 (2021) 118998. https://doi.org/10.1016/j.bbamcr.2021.118998.

[140] V.L. Hewitt, L. Miller-Fleming, M.J. Twyning, S. Andreazza, F. Mattedi, J. Prudent, F. Polleux, A. Vagnoni, A.J. Whitworth, Decreasing pdzd8-mediated mito-ER contacts improves organismal fitness and mitigates Aβ42 toxicity, Life Sci. Alliance. 5 (2022) e202201531. https://doi.org/10.26508/lsa.202201531.

[141] D. Lim, G. Dematteis, L. Tapella, A.A. Genazzani, T. Calì, M. Brini, A. Verkhratsky, Ca^{2+} handling at the mitochondria-ER contact sites in neurodegeneration, Cell Calcium. 98 (2021) 102453. https://doi.org/10.1016/j.ceca.2021.102453.

[142] T. Thoudam, C.M. Ha, J. Leem, D. Chanda, J.S. Park, H.J. Kim, J.H. Jeon, Y.K. Choi, S. Liangpunsakul, Y.H. Huh, T.H. Kwon, K.G. Park, R.A. Harris, K.S. Park, H.W. Rhee, I.K. Lee, PDK4 augments ER–mitochondria contact to dampen skeletal muscle insulin signaling during obesity, Diabetes. 68 (2019) 571–586. https://doi.org/10.2337/db18-0363.

[143] C.H. Wang, Y.H. Wei, Role of mitochondrial dysfunction and dysregulation of Ca^{2+} homeostasis in the pathophysiology of insulin resistance and type 2 diabetes, J. Biomed. Sci. 24 (2017) 70. https://doi.org/10.1186/s12929-017-0375-3.

[144] L. Pietrangelo, A. D'Incecco, A. Ainbinder, A. Michelucci, H. Kern, R.T. Dirksen, S. Boncompagni, F. Protasi, Age-dependent uncoupling of mitochondria from Ca^{2+} release units in skeletal muscle, Oncotarget. 6 (2015) 35358–35371. https://doi.org/10.18632/oncotarget.6139.

[145] J.F. Gill, J. Delezie, G. Santos, S. McGuirk, S. Schnyder, S. Frank, M. Rausch, J. St-Pierre, C. Handschin, Peroxisome proliferator-activated receptor γ coactivator 1α regulates

mitochondrial calcium homeostasis, sarcoplasmic reticulum stress, and cell death to mitigate skeletal muscle aging, Aging Cell. 18 (2019) e12993. https://doi.org/10.1111/acel.12993.

[146] A. del Campo, I. Contreras-Hernández, M. Castro-Sepúlveda, C.A. Campos, R. Figueroa, M.F. Tevy, V. Eisner, M. Casas, E. Jaimovich, Muscle function decline and mitochondria changes in middle age precede sarcopenia in mice, Aging (Albany. NY). 10 (2018) 34–55. https://doi.org/10.18632/aging.101358.

[147] M. Pauly, C. Angebault-Prouteau, H. Dridi, C. Notarnicola, V. Scheuermann, A. Lacampagne, S. Matecki, J. Fauconnier, ER stress disturbs SR/ER-mitochondria Ca^{2+} transfer: Implications in Duchenne muscular dystrophy, Biochim. Biophys. Acta - Mol. Basis Dis. 1863 (2017) 2229–2239. https://doi.org/10.1016/j.bbadis.2017.06.009.

[148] S. Zampieri, C. Mammucari, V. Romanello, L. Barberi, L. Pietrangelo, A. Fusella, S. Mosole, G. Gherardi, C. Höfer, S. Löfler, N. Sarabon, J. Cvecka, M. Krenn, U. Carraro, H. Kern, F. Protasi, A. Musarò, M. Sandri, R. Rizzuto, Physical exercise in aging human skeletal muscle increases mitochondrial calcium uniporter expression levels and affects mitochondria dynamics, Physiol. Rep. 4 (2016) e13005. https://doi.org/10.14814/phy2.13005.

[149] S.S. Zhang, S. Zhou, Z.J. Crowley-Mchattan, R.Y. Wang, J.P. Li, A review of the role of endo/sarcoplasmic reticulum-mitochondria Ca^{2+} transport in diseases and skeletal muscle function, Int. J. Environ. Res. Public Health. 18 (2021) 3874. https://doi.org/10.3390/ijerph18083874.

[150] Y. Lv, Y. Li, D. Zhang, A. Zhang, W. Guo, S. Zhu, HMGB1-induced asthmatic airway inflammation through GRP75-mediated enhancement of ER-mitochondrial Ca^{2+} transfer and ROS increased, J. Cell. Biochem. 119 (2018) 4205–4215. https://doi.org/10.1002/jcb.26653.

[151] J. Li, F. Qi, H. Su, C. Zhang, Q. Zhang, Y. Chen, P. Chen, L. Su, Y. Chen, Y. Yang, Z. Chen, S. Zhang, GRP75-faciliated mitochondria-associated ER membrane (MAM) integrity controls cisplatin-resistance in ovarian cancer patients, Int. J. Biol. Sci. 18 (2022) 2914–2931. https://doi.org/10.7150/ijbs.71571.

[152] G. Gherardi, H. Monticelli, R. Rizzuto, C. Mammucari, The mitochondrial Ca^{2+} uptake and the fine-tuning of aerobic metabolism, Front. Physiol. 11 (2020) 554904. https://doi.org/10.3389/fphys.2020.554904.

[153] R.M. Denton, Regulation of mitochondrial dehydrogenases by calcium ions, Biochim. Biophys. Acta - Bioenerg. 1787 (2009) 1309–1316. https://doi.org/10.1016/j.bbabio.2009.01.005.

[154] C. Cárdenas, R.A. Miller, I. Smith, T. Bui, J. Molgó, M. Müller, H. Vais, K.H. Cheung, J. Yang, I. Parker, C.B. Thompson, M.J. Birnbaum, K.R. Hallows, J.K. Foskett, Essential regulation of cell bioenergetics by constitutive $InsP_3$ receptor Ca^{2+} transfer to mitochondria, Cell. 142 (2010) 270–283. https://doi.org/10.1016/j.cell.2010.06.007.

[155] M.J. MacDonald, L.J. Brown, Calcium activation of mitochondrial glycerol phosphate dehydrogenase restudied, Arch. Biochem. Biophys. 326 (1996) 79–84. https://doi.org/10.1006/abbi.1996.0049.

[156] T. Mráček, Z. Drahota, J. Houštěk, The function and the role of the mitochondrial glycerol-3-phosphate dehydrogenase in mammalian tissues, Biochim. Biophys. Acta - Bioenerg. 1827 (2013) 401–410. https://doi.org/10.1016/j.bbabio.2012.11.014.

[157] G.A. Rutter, W.F. Pralong, C.B. Wollheim, Regulation of mitochondrial glycerol-phosphate dehydrogenase by Ca^{2+} within electropermeabilized insulin-secreting cells (INS-1), BBA - Mol. Cell Res. 1175 (1992) 107–113. https://doi.org/10.1016/0167-4889(92)90016-5.

[158] R.M. Denton, J.G. McCormack, G.A. Rutter, P. Burnett, N.J. Edgell, S.K. Moule, T.A. Diggle, The hormonal regulation of pyruvate dehydrogenase complex, Adv. Enzyme Regul. 36 (1996) 183–198. https://doi.org/10.1016/0065-2571(95)00020-8.

[159] Y. Hiromasa, T. Fujisawa, Y. Aso, T.E. Roche, Organization of the cores of the mammalian pyruvate dehydrogenase complex formed by E2 and E2 plus the E3-binding protein and their capacities to bind the E1 and E3 components, J. Biol. Chem. 279 (2004) 6921–6933. https://doi.org/10.1074/jbc.M308172200.

[160] W.M. Teague, F.H. Pettit, T.L. Wu, S.R. Silberman, L.J. Reed, Purification and properties of pyruvate dehydrogenase phosphatase from bovine heart and kidney, Biochemistry. 21 (1982) 5585–5592. https://doi.org/10.1021/bi00265a031.

[161] A. Turkan, Y. Hiromasa, T.E. Roche, Formation of a complex of the catalytic subunit of pyruvate dehydrogenase phosphatase isoform 1 (PDP1c) and the L2 domain forms a Ca^{2+} binding site and captures PDP1c as a monomer, Biochemistry. 43 (2004) 15073–15085. https://doi.org/10.1021/bi048901y.

[162] D.G. Vassylyev, J. Symersky, Crystal structure of pyruvate dehydrogenase phosphatase 1 and its functional implications, J. Mol. Biol. 370 (2007) 417–426. https://doi.org/10.1016/j.jmb.2007.05.002.

[163] B.J. Nichols, L. Hall, A.C.F. Perry, R.M. Denton, Molecular cloning and deduced amino acid

sequences of the γ-subunits of rat and monkey NAD^+-isocitrate dehydrogenases, Biochem. J. 295 (1993) 347–350. https://doi.org/10.1042/bj2950347.

[164] B.J. Nichols, A.C.F. Perry, L. Hall, R.M. Denton, Molecular cloning and deduced amino acid sequences of the α- and β-subunits of mammalian NAD^+-isocitrate dehydrogenase, Biochem. J. 310 (1995) 917–922. https://doi.org/10.1042/bj3100917.

[165] G.A. Rutter, R.M. Denton, Regulation of NAD^+-linked isocitrate dehydrogenase and 2-oxoglutarate dehydrogenase by Ca^{2+} ions within toluene-permeabilized rat heart mitochondria. Interactions with regulation by adenine nucleotides and NADH/NAD^+ ratios, Biochem. J. 252 (1988) 181–189. https://doi.org/10.1042/bj2520181.

[166] G.A. Rutter, R.M. Denton, The binding of Ca^{2+} ions to pig heart NAD^+-isocitrate dehydrogenase and the 2-oxoglutarate dehydrogenase complex, Biochem. J. 263 (1989) 453–462. https://doi.org/10.1042/bj2630453.

[167] R.M. Denton, D.A. Richards, J.G. Chin, Calcium ions and the regulation of NAD^+-linked isocitrate dehydrogenase from the mitochondria of rat heart and other tissues, Biochem. J. 176 (1978) 899–906. https://doi.org/10.1042/bj1760899.

[168] J.G. McCormack, R.M. Denton, The effects of calcium ions and adenine nucleotides on the activity of pig heart 2-oxoglutarate dehydrogenase complex, Biochem. J. 180 (1979) 533–544. https://doi.org/10.1042/bj1800533.

[169] V.B. Lawlis, T.E. Roche, Regulation of bovine kidney α-ketoglutarate dehydrogenase complex by calcium ion and adenine nucleotides, effects on $S_{0.5}$ for α-ketoglutarate, Biochemistry. 20 (1981) 2512–2518. https://doi.org/10.1021/bi00512a023.

[170] C.T. Armstrong, J.L.R. Anderson, R.M. Denton, Studies on the regulation of the human E1 subunit of the 2-oxoglutarate dehydrogenase complex, including the identification of a novel calcium-binding site, Biochem. J. 459 (2014) 369–381. https://doi.org/10.1042/BJ20131664.

[171] R.M. Denton, T.J. Pullen, C.T. Armstrong, K.J. Heesom, G.A. Rutter, Calcium-insensitive splice variants of mammalian E1 subunit of 2-oxoglutarate dehydrogenase complex with tissue-specific patterns of expression, Biochem. J. 473 (2016) 1165–1178. https://doi.org/10.1042/BCJ20160135.

[172] B. Glancy, W.T. Willis, D.J. Chess, R.S. Balaban, Effect of calcium on the oxidative phosphorylation cascade in skeletal muscle mitochondria, Biochemistry. 52 (2013) 2793–2809. https://doi.org/10.1021/bi3015983.

[173] P.R. Territo, V.K. Mootha, S.A. French, R.S. Balaban, Ca^{2+} activation of heart mitochondrial oxidative phosphorylation: Role of the F_0/F_1-ATPase, Am. J. Physiol. - Cell Physiol. 278 (2000) C423-435. https://doi.org/10.1152/ajpcell.2000.278.2.c423.

[174] F.M. Lasorsa, P. Pinton, L. Palmieri, G. Fiermonte, R. Rizzuto, F. Palmieri, Recombinant expression of the Ca^{2+}-sensitive aspartate/ glutamate carrier increases mitochondrial ATP production in agonist-stimulated Chinese hamster ovary cells, J. Biol. Chem. 278 (2003) 38686–38692. https://doi.org/10.1074/jbc.M304988200.

[175] L. Palmieri, B. Pardo, F.M. Lasorsa, A. Del Arco, K. Kobayashi, M. Iijima, M.J. Runswick, J.E. Walker, T. Saheki, J. Satrústegui, F. Palmieri, Citrin and aralar1 are Ca^{2+}-stimulated aspartate/glutamate transporters in mitochondria, EMBO J. 20 (2001) 5060–5069. https://doi.org/10.1093/emboj/20.18.5060.

[176] A. Del Arco, J. Satrústegui, Molecular cloning of aralar, a new member of the mitochondrial carrier superfamily that binds calcium and is present in human muscle and brain, J. Biol. Chem. 273 (1998) 23327–23334. https://doi.org/10.1074/jbc.273.36.23327.

[177] K. Kobayashi, D.S. Sinasac, M. Iijima, A.P. Boright, L. Begum, J.R. Lee, T. Yasuda, S. Ikeda, R. Hirano, H. Terazono, M.A. Crackower, I. Kondo, L.C. Tsui, S.W. Scherer, T. Saheki, The gene mutated in adult-onset type II citrullinaemia encodes a putative mitochondrial carrier protein, Nat. Genet. 22 (1999) 159–163. https://doi.org/10.1038/9667.

[178] B. Pardo, L. Contreras, A. Serrano, M. Ramos, K. Kobayashi, M. Iijima, T. Saheki, J. Satrústegui, Essential role of aralar in the transduction of small Ca^{2+} signals to neuronal mitochondria, J. Biol. Chem. 281 (2006) 1039–1047. https://doi.org/10.1074/jbc.M507270200.

[179] L. Contreras, P. Gomez-Puertas, M. Iijima, K. Kobayashi, T. Saheki, J. Satrústegui, Ca^{2+} activation kinetics of the two aspartate-glutamate mitochondrial carriers, aralar and citrin: Role in the heart malate-aspartate NADH shuttle, J. Biol. Chem. 282 (2007) 7098–7106. https://doi.org/10.1074/jbc.M610491200.

[180] R.P. Anunciado-Koza, J. Zhang, J. Ukropec, S. Bajpeyi, R.A. Koza, R.C. Rogers, W.T. Cefalu, R.L. Mynatt, L.P. Kozak, Inactivation of the mitochondrial carrier SLC25A25 (ATP-Mg^{2+}/P_i transporter) reduces physical endurance and metabolic efficiency in mice, J. Biol. Chem. 286 (2011) 11659–11671. https://doi.org/10.1074/jbc.M110.203000.

[181] M.T. Nosek, D.T. Dransfield, J.R. Aprille, Calcium stimulates ATP-Mg/P_i carrier activity in rat liver mitochondria, J. Biol. Chem. 265 (1990) 8444–8450. https://doi.org/10.1016/s0021-

9258(19)38908-2.

[182] A. Del Arco, J. Satrústegui, Identification of a novel human subfamily of mitochondrial carriers with calcium-binding domains, J. Biol. Chem. 279 (2004) 24701–24713. https://doi.org/10.1074/jbc.M401417200.

[183] Q. Yang, S. Brüschweiler, J.J. Chou, A self-sequestered calmodulin-like Ca^{2+} sensor of mitochondrial SCaMC carrier and its implication to Ca^{2+}-dependent ATP-Mg/P_i transport, Structure. 22 (2014) 209–217. https://doi.org/10.1016/j.str.2013.10.018.

[184] S.P.D. Harborne, J.J. Ruprecht, E.R.S. Kunji, Calcium-induced conformational changes in the regulatory domain of the human mitochondrial ATP-Mg/P_i carrier, Biochim. Biophys. Acta - Bioenerg. 1847 (2015) 1245–1253. https://doi.org/10.1016/j.bbabio.2015.07.002.

[185] S. García-Catalán, L. González-Moreno, A. del Arco, Ca^{2+}-regulated mitochondrial carriers of ATP-Mg^{2+}/P_i: Evolutionary insights in protozoans, Biochim. Biophys. Acta - Mol. Cell Res. 1868 (2021) 119038. https://doi.org/10.1016/j.bbamcr.2021.119038.

[186] C. Cárdenas, J.K. Foskett, Mitochondrial Ca^{2+} signals in autophagy, Cell Calcium. 52 (2012) 44-51. https://doi.org/10.1016/j.ceca.2012.03.001.

[187] G. Szabadkai, A.M. Simoni, K. Bianchi, D. De Stefani, S. Leo, M.R. Wieckowski, R. Rizzuto, Mitochondrial dynamics and Ca^{2+} signaling, Biochim. Biophys. Acta - Mol. Cell Res. 1763 (2006) 442–449. https://doi.org/10.1016/j.bbamcr.2006.04.002.

[188] Y. Ohshima, N. Takata, M. Suzuki-Karasaki, Y. Yoshida, Y. Tokuhashi, Y. Suzuki-Karasaki, Disrupting mitochondrial Ca^{2+} homeostasis causes tumor-selective TRAIL sensitization through mitochondrial network abnormalities, Int. J. Oncol. 51 (2017) 1146–1158. https://doi.org/10.3892/ijo.2017.4096.

[189] K.T. Chang, R.F. Niescier, K.T. Min, Mitochondrial matrix Ca^{2+} as an intrinsic signal regulating mitochondrial motility in axons, Proc. Natl. Acad. Sci. U. S. A. 108 (2011) 15456–15461. https://doi.org/10.1073/pnas.1106862108.

[190] S. Marchi, S. Patergnani, S. Missiroli, G. Morciano, A. Rimessi, M.R. Wieckowski, C. Giorgi, P. Pinton, Mitochondrial and endoplasmic reticulum calcium homeostasis and cell death, Cell Calcium. 69 (2018) 62–72. https://doi.org/10.1016/J.CECA.2017.05.003.

[191] K.W. Kinnally, P.M. Peixoto, S.Y. Ryu, L.M. Dejean, Is mPTP the gatekeeper for necrosis, apoptosis, or both?, Biochim. Biophys. Acta - Mol. Cell Res. 1813 (2011) 616–622. https://doi.org/10.1016/j.bbamcr.2010.09.013.

[192] M.A. Smith, R.G. Schnellmann, Calpains, mitochondria, and apoptosis, Cardiovasc. Res. 96 (2012) 32–37. https://doi.org/10.1093/cvr/cvs163.

[193] H.K. Baumgartner, J. V. Gerasimenko, C. Thorne, P. Ferdek, T. Pozzan, A. V. Tepikin, O.H. Petersen, R. Sutton, A.J.M. Watson, O. V. Gerasimenko, Calcium elevation in mitochondria is the main Ca^{2+} requirement for mitochondrial permeability transition pore (mPTP) opening, J. Biol. Chem. 284 (2009) 20796–20803. https://doi.org/10.1074/jbc.M109.025353.

[194] V. Giorgio, V. Burchell, M. Schiavone, C. Bassot, G. Minervini, V. Petronilli, F. Argenton, M. Forte, S. Tosatto, G. Lippe, P. Bernardi, Ca^{2+} binding to F-ATP synthase β subunit triggers the mitochondrial permeability transition, EMBO Rep. 18 (2017) 1065–1076. https://doi.org/10.15252/embr.201643354.

[195] R. Wong, C. Steenbergen, E. Murphy, Mitochondrial permeability transition pore and calcium handling, Methods Mol. Biol. 810 (2012) 235–242. https://doi.org/10.1007/978-1-61779-382-0_15.

[196] M.S. Hwang, C.T. Schwall, E. Pazarentzos, C. Datler, N.N. Alder, S. Grimm, Mitochondrial Ca^{2+} influx targets cardiolipin to disintegrate respiratory chain complex II for cell death induction, Cell Death Differ. 21 (2014) 1733–1745. https://doi.org/10.1038/cdd.2014.84.

[197] M. Bonora, C. Giorgi, P. Pinton, Molecular mechanisms and consequences of mitochondrial permeability transition, Nat. Rev. Mol. Cell Biol. 23 (2022) 266–285. https://doi.org/10.1038/s41580-021-00433-y.

[198] A.P. Halestrap, What is the mitochondrial permeability transition pore?, J. Mol. Cell. Cardiol. 46 (2009) 821–831. https://doi.org/10.1016/j.yjmcc.2009.02.021.

[199] J.Q. Kwong, J.D. Molkentin, Physiological and pathological roles of the mitochondrial permeability transition pore in the heart, Cell Metab. 21 (2015) 206–214. https://doi.org/10.1016/j.cmet.2014.12.001.

[200] M. Carraro, P. Bernardi, The mitochondrial permeability transition pore in Ca^{2+} homeostasis., Cell Calcium. 111 (2023) 102719.

[201] K.N. Alavian, G. Beutner, E. Lazrove, S. Sacchetti, H.A. Park, P. Licznerski, H. Li, P. Nabili, K. Hockensmith, M. Graham, G.A. Porter, E.A. Jonas, An uncoupling channel within the c-subunit ring of the F_1F_O ATP synthase is the mitochondrial permeability transition pore, Proc. Natl. Acad. Sci. U. S. A. 111 (2014) 10580–10585. https://doi.org/10.1073/pnas.1401591111.

[202] V. Giorgio, S. Von Stockum, M. Antoniel, A. Fabbro, F. Fogolari, M. Forte, G.D. Glick, V.

Petronilli, M. Zoratti, I. Szabó, G. Lippe, P. Bernardi, Dimers of mitochondrial ATP synthase form the permeability transition pore, Proc. Natl. Acad. Sci. U. S. A. 110 (2013) 5887–5892. https://doi.org/10.1073/pnas.1217823110.

[203] M. Bonora, C. Morganti, G. Morciano, G. Pedriali, M. Lebiedzinska-Arciszewska, G. Aquila, C. Giorgi, P. Rizzo, G. Campo, R. Ferrari, G. Kroemer, M.R. Wieckowski, L. Galluzzi, P. Pinton, Mitochondrial permeability transition involves dissociation of F_1F_0 ATP synthase dimers and C-ring conformation, EMBO Rep. 18 (2017) 1077–1089. https://doi.org/10.15252/embr.201643602.

[204] M. Bonora, A. Bononi, E. De Marchi, C. Giorgi, M. Lebiedzinska, S. Marchi, S. Patergnani, A. Rimessi, J.M. Suski, A. Wojtala, M.R. Wieckowski, G. Kroemer, L. Galluzzi, P. Pinton, Role of the c subunit of the F_O ATP synthase in mitochondrial permeability transition, Cell Cycle. 12 (2013) 674–683. https://doi.org/10.4161/cc.23599.

[205] V. Giorgio, E. Bisetto, M.E. Soriano, F. Dabbeni-Sala, E. Basso, V. Petronilli, M.A. Forte, P. Bernardi, G. Lippe, Cyclophilin D modulates mitochondrial F_0F_1-ATP synthase by interacting with the lateral stalk of the complex, J. Biol. Chem. 284 (2009) 33982–33988. https://doi.org/10.1074/jbc.M109.020115.

[206] G. Cannino, A. Urbani, M. Gaspari, M. Varano, A. Negro, A. Filippi, F. Ciscato, I. Masgras, C. Gerle, E. Tibaldi, A.M. Brunati, G. Colombo, G. Lippe, P. Bernardi, A. Rasola, The mitochondrial chaperone TRAP1 regulates F-ATP synthase channel formation, Cell Death Differ. 29 (2022) 2335–2346. https://doi.org/10.1038/s41418-022-01020-0.

[207] J. He, H.C. Ford, J. Carroll, S. Ding, I.M. Fearnley, J.E. Walker, Persistence of the mitochondrial permeability transition in the absence of subunit c of human ATP synthase, Proc. Natl. Acad. Sci. U. S. A. 114 (2017) 3409–3414. https://doi.org/10.1073/pnas.1702357114.

[208] N. Mnatsakanyan, M.C. Llaguno, Y. Yang, Y. Yan, J. Weber, F.J. Sigworth, E.A. Jonas, A mitochondrial megachannel resides in monomeric F_1F_O ATP synthase, Nat. Commun. 10 (2019) 5823. https://doi.org/10.1038/s41467-019-13766-2.

[209] J. Karch, M.J. Bround, H. Khalil, M.A. Sargent, N. Latchman, N. Terada, P.M. Peixoto, J.D. Molkentin, Inhibition of mitochondrial permeability transition by deletion of the ANT family and CypD, Sci. Adv. 5 (2019) 4597. https://doi.org/10.1126/sciadv.aaw4597.

[210] M.A. Neginskaya, M.E. Solesio, E. V. Berezhnaya, G.F. Amodeo, N. Mnatsakanyan, E.A. Jonas, E. V. Pavlov, ATP synthase C-subunit-deficient mitochondria have a small cyclosporine A-

sensitive channel, but lack the permeability transition pore, Cell Rep. 26 (2019) 11-17.e2. https://doi.org/10.1016/j.celrep.2018.12.033.

[211] H. Ivanova, T. Vervliet, L. Missiaen, J.B. Parys, H. De Smedt, G. Bultynck, Inositol 1,4,5-trisphosphate receptor-isoform diversity in cell death and survival, Biochim. Biophys. Acta - Mol. Cell Res. 1843 (2014) 2164–2183. https://doi.org/10.1016/J.BBAMCR.2014.03.007.

[212] T. Vervloessem, D.I. Yule, G. Bultynck, J.B. Parys, The type 2 inositol 1,4,5-trisphosphate receptor, emerging functions for an intriguing Ca^{2+}-release channel, Biochim. Biophys. Acta - Mol. Cell Res. 1853 (2014) 1992–2005. https://doi.org/10.1016/j.bbamcr.2014.12.006.

[213] N. Rosa, F. Sneyers, J.B. Parys, G. Bultynck, Type 3 IP3 receptors: The chameleon in cancer, Int. Rev. Cell Mol. Biol. 351 (2020) 101–148. https://doi.org/10.1016/bs.ircmb.2020.02.003.

[214] H.A. Smith, N.B. Thillaiappan, A.M. Rossi, IP_3 receptors: an “elementary” journey from structure to signals., Cell Calcium. in press (2023).

[215] D.A. Gomes, M. Thompson, N.C. Souto, T.S. Goes, A.M. Goes, M.A. Rodrigues, M. V. Gomez, M.H. Nathanson, M.F. Leite, The type III inositol 1,4,5-trisphosphate receptor preferentially transmits apoptotic Ca^{2+} signals into mitochondria, J. Biol. Chem. 280 (2005) 40892–40900. https://doi.org/10.1074/JBC.M506623200.

[216] S. Marchi, M. Marinello, A. Bononi, M. Bonora, C. Giorgi, A. Rimessi, P. Pinton, Selective modulation of subtype III IP_3R by Akt regulates ER Ca^{2+} release and apoptosis, Cell Death Dis. 3 (2012) 304. https://doi.org/10.1038/CDDIS.2012.45.

[217] T. Jayaraman, A.R. Marks, T cells deficient in inositol 1,4,5-trisphosphate receptor are resistant to apoptosis, Mol. Cell. Biol. 17 (1997) 3005–3012. https://doi.org/10.1128/mcb.17.6.3005.

[218] A. Bononi, M. Bonora, S. Marchi, S. Missiroli, F. Poletti, C. Giorgi, P.P. Pandolfi, P. Pinton, Identification of PTEN at the ER and MAMs and its regulation of Ca^{2+} signaling and apoptosis in a protein phosphatase-dependent manner, Cell Death Differ. 20 (2013) 1631–1643. https://doi.org/10.1038/cdd.2013.77.

[219] V. Álvarez-Garcia, Y. Tawil, H.M. Wise, N.R. Leslie, Mechanisms of PTEN loss in cancer: It’s all about diversity, Semin. Cancer Biol. 59 (2019) 66–79. https://doi.org/10.1016/j.semcancer.2019.02.001.

[220] M. Bittremieux, J.B. Parys, P. Pinton, G. Bultynck, ER functions of oncogenes and tumor suppressors: Modulators of intracellular Ca^{2+} signaling, Biochim. Biophys. Acta - Mol. Cell Res.

1863 (2016) 1364–1378. https://doi.org/10.1016/j.bbamcr.2016.01.002.

[221] J.B. Parys, G. Bultynck, T. Vervliet, IP_3 receptor biology and endoplasmic reticulum calcium dynamics in cancer, Prog. Mol. Subcell. Biol. 59 (2021) 215–237. https://doi.org/10.1007/978-3-030-67696-4_11.

[222] F. Ciscato, R. Filadi, I. Masgras, M. Pizzi, O. Marin, N. Damiano, P. Pizzo, A. Gori, F. Frezzato, F. Chiara, L. Trentin, P. Bernardi, A. Rasola, Hexokinase 2 displacement from mitochondria-associated membranes prompts Ca^{2+}-dependent death of cancer cells, EMBO Rep. 21 (2020) e49117. https://doi.org/10.15252/embr.201949117.

[223] A.R. Lavik, K.S. McColl, F.O. Lemos, M. Kerkhofs, F. Zhong, M. Harr, D. Schlatzer, K. Hamada, K. Mikoshiba, F. Crea, G. Bultynck, M.D. Bootman, J.B. Parys, C.W. Distelhorst, A non-canonical role for pyruvate kinase M2 as a functional modulator of Ca^{2+} signalling through IP_3 receptors, Biochim. Biophys. Acta - Mol. Cell Res. 1869 (2022) 119206. https://doi.org/10.1016/j.bbamcr.2021.119206.

[224] J. Kale, E.J. Osterlund, D.W. Andrews, BCL-2 family proteins: Changing partners in the dance towards death, Cell Death Differ. 25 (2018) 65–80. https://doi.org/10.1038/cdd.2017.186.

[225] P. Vandenabeele, G. Bultynck, S.N. Savvides, Pore-forming proteins as drivers of membrane permeabilization in cell death pathways, Nat. Rev. Mol. Cell Biol. (2022). https://doi.org/10.1038/s41580-022-00564-w.

[226] T. Vervliet, J. Parys, G. Bultynck, Bcl-2 proteins and calcium signaling: complexity beneath the surface, Oncogene. 35 (2016) 5079–5092. https://doi.org/10.1038/ONC.2016.31.

[227] H. Ivanova, T. Vervliet, G. Monaco, L.E. Terry, N. Rosa, M.R. Baker, J.B. Parys, I.I. Serysheva, D.I. Yule, G. Bultynck, Bcl-2-protein family as modulators of IP_3 receptors and other organellar Ca^{2+} channels, Cold Spring Harb. Perspect. Biol. 12 (2020) a035089. https://doi.org/10.1101/CSHPERSPECT.A035089.

[228] N. Rosa, F. Speelman-Rooms, J.B. Parys, G. Bultynck, Modulation of Ca^{2+} signaling by antiapoptotic Bcl-2 versus Bcl-xL: From molecular mechanisms to relevance for cancer cell survival, Biochim. Biophys. Acta. Rev. Cancer. 1877 (2022) 188791. https://doi.org/10.1016/j.bbcan.2022.188791.

[229] Y.P. Rong, G. Bultynck, A.S. Aromolaran, F. Zhong, J.B. Parys, H. De Smedt, G.A. Mignery, H.L. Roderick, M.D. Bootman, C.W. Distelhorst, The BH4 domain of Bcl-2 inhibits ER calcium release and apoptosis by binding the regulatory and coupling domain of the IP3 receptor,

Proc. Natl. Acad. Sci. U. S. A. 106 (2009) 14397–14402. https://doi.org/10.1073/pnas.0907555106.

[230] G. Monaco, E. Decrock, H. Akl, R. Ponsaerts, T. Vervliet, T. Luyten, M. De Maeyer, L. Missiaen, C.W. Distelhorst, H. De Smedt, J.B. Parys, L. Leybaert, G. Bultynck, Selective regulation of IP_3-receptor-mediated Ca^{2+} signaling and apoptosis by the BH4 domain of Bcl-2 versus Bcl-Xl, Cell Death Differ. 19 (2012) 295–309. https://doi.org/10.1038/cdd.2011.97.

[231] G. Monaco, M. Beckers, H. Ivanova, L. Missiaen, J.B. Parys, H. De Smedt, G. Bultynck, Profiling of the Bcl-2/Bcl-X_L-binding sites on type 1 IP_3 receptor, Biochem. Biophys. Res. Commun. 428 (2012) 31–35. https://doi.org/10.1016/J.BBRC.2012.10.002.

[232] H. Ivanova, L.E. Wagner, A. Tanimura, E. Vandermarliere, T. Luyten, K. Welkenhuyzen, K.J. Alzayady, L. Wang, K. Hamada, K. Mikoshiba, H. De Smedt, L. Martens, D.I. Yule, J.B. Parys, G. Bultynck, Bcl-2 and IP_3 compete for the ligand-binding domain of IP_3Rs modulating Ca^{2+} signaling output, Cell. Mol. Life Sci. 76 (2019) 3843–3859. https://doi.org/10.1007/S00018-019-03091-8.

[233] J. Yang, H. Vais, W. Gu, J.K. Foskett, A.R. Marks, Biphasic regulation of $InsP_3$ receptor gating by dual Ca^{2+} release channel BH3-like domains mediates Bcl-xL control of cell viability, Proc. Natl. Acad. Sci. U. S. A. 113 (2016) E1953–E1962. https://doi.org/10.1073/pnas.1517935113.

[234] P.J. Sung, F.D. Tsai, H. Vais, H. Court, J. Yang, N. Fehrenbacher, J.K. Foskett, M.R. Philips, Phosphorylated K-Ras limits cell survival by blocking Bcl-xL sensitization of inositol trisphosphate receptors, Proc. Natl. Acad. Sci. U. S. A. 110 (2013) 20593–20598. https://doi.org/10.1073/pnas.1306431110.

[235] N. Rosa, H. Ivanova, L.E. Wagner, J. Kale, R. La Rovere, K. Welkenhuyzen, N. Louros, S. Karamanou, V. Shabardina, I. Lemmens, E. Vandermarliere, K. Hamada, H. Ando, F. Rousseau, J. Schymkowitz, J. Tavernier, K. Mikoshiba, A. Economou, D.W. Andrews, J.B. Parys, D.I. Yule, G. Bultynck, Bcl-xL acts as an inhibitor of IP_3R channels, thereby antagonizing Ca^{2+}-driven apoptosis, Cell Death Differ. 29 (2022) 788–805. https://doi.org/10.1038/s41418-021-00894-w.

[236] J.J. Schulman, F.A. Wright, T. Kaufmann, R.J.H. Wojcikiewicz, 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 (2013) 25340–25349. https://doi.org/10.1074/jbc.M113.496570.

[237] J.J. Schulman, F.A. Wright, X. Han, E.J. Zluhan, L.M. Szczesniak, R.J.H. Wojcikiewicz, The stability and expression level of Bok are governed by binding to inositol 1,4,5-trisphosphate receptors, J. Biol. Chem. 291 (2016) 11820–11828. https://doi.org/10.1074/jbc.M115.711242.

[238] F. Llambi, Y.M. Wang, B. Victor, M. Yang, D.M. Schneider, S. Gingras, M.J. Parsons, J.H. Zheng, S.A. Brown, S. Pelletier, T. Moldoveanu, T. Chen, D.R. Green, BOK is a non-canonical BCL-2 family effector of apoptosis regulated by ER-associated degradation, Cell. 165 (2016) 421–433. https://doi.org/10.1016/j.cell.2016.02.026.

[239] M.A. Carpio, R.E. Means, A.L. Brill, A. Sainz, B.E. Ehrlich, S.G. Katz, BOK controls apoptosis by Ca^{2+} transfer through ER-mitochondrial contact sites, Cell Rep. 34 (2021) 108827. https://doi.org/10.1016/j.celrep.2021.108827.

[240] F. Zhong, M.W. Harr, G. Bultynck, G. Monaco, J.B. Parys, H. De Smedt, Y.P. Rong, J.K. Molitoris, M. Lam, C. Ryder, S. Matsuyama, C.W. Distelhorst, Induction of Ca^{2+}-driven apoptosis in chronic lymphocytic leukemia cells by peptide-mediated disruption of Bcl-2-IP_3 receptor interaction, Blood. 117 (2011) 2924–2934. https://doi.org/10.1182/blood-2010-09-307405.

[241] H. Akl, G. Monaco, R. La Rovere, K. Welkenhuyzen, S. Kiviluoto, T. Vervliet, J. Molgó, C.W. Distelhorst, L. Missiaen, K. Mikoshiba, J.B. Parys, H. De Smedt, G. Bultynck, IP_3R2 levels dictate the apoptotic sensitivity of diffuse large B-cell lymphoma cells to an IP_3R-derived peptide targeting the BH4 domain of Bcl-2, Cell Death Dis. 4 (2013) e632. https://doi.org/10.1038/cddis.2013.140.

[242] A.R. Lavik, F. Zhong, M.J. Chang, E. Greenberg, Y. Choudhary, M.R. Smith, K.S. McColl, J. Pink, F.J. Reu, S. Matsuyama, C.W. Distelhorst, A synthetic peptide targeting the BH4 domain of Bcl-2 induces apoptosis in multiple myeloma and follicular lymphoma cells alone or in combination with agents targeting the BH3-binding pocket of Bcl-2, Oncotarget. 6 (2015) 27388–27402. https://doi.org/10.18632/oncotarget.4489.

[243] M. Kerkhofs, R. La Rovere, K. Welkenhuysen, A. Janssens, P. Vandenberghe, M. Madesh, J.B. Parys, G. Bultynck, BIRD-2, a BH4-domain-targeting peptide of Bcl-2, provokes Bax/Bak-independent cell death in B-cell cancers through mitochondrial Ca^{2+}-dependent mPTP opening, Cell Calcium. 94 (2021) 102333. https://doi.org/10.1016/j.ceca.2020.102333.

[244] T. Vervloessem, M. Kerkhofs, R.M. La Rovere, F. Sneyers, J.B. Parys, G. Bultynck, Bcl-2 inhibitors as anti-cancer therapeutics: The impact of and on calcium signaling, Cell Calcium. 70 (2018) 102–116. https://doi.org/10.1016/j.ceca.2017.05.014.

[245] Q. Xie, Y. Xu, W. Gao, Y. Zhang, J. Su, Y. Liu, Y. Guo, M. Dou, K. Hu, L. Sun, TAT-fused IP_3R-derived peptide enhances cisplatin sensitivity of ovarian cancer cells by increasing ER Ca^{2+} release, Int. J. Mol. Med. 41 (2018) 809–817. https://doi.org/10.3892/ijmm.2017.3260.

[246] G. Monaco, E. Decrock, N. Arbel, A.R. Van Vliet, R.M. La Rovere, H. De Smedt, J.B. Parys, P. Agostinis, L. Leybaert, V. Shoshan-Barmatz, G. Bultynck, The BH4 domain of anti-apoptotic Bcl-XL, but not that of the related Bcl-2, limits the voltage-dependent anion channel 1 (VDAC1)-mediated transfer of pro-apoptotic Ca^{2+} signals to mitochondria, J. Biol. Chem. 290 (2015) 9150–9161. https://doi.org/10.1074/jbc.M114.622514.

[247] S. Marchi, P. Pinton, The mitochondrial calcium uniporter complex: Molecular components, structure and physiopathological implications, J. Physiol. 592 (2014) 829–839. https://doi.org/10.1113/jphysiol.2013.268235.

[248] C. Mammucari, G. Gherardi, R. Rizzuto, Structure, activity regulation, and role of the mitochondrial calcium uniporter in health and disease, Front. Oncol. 7 (2017) 139. https://doi.org/10.3389/fonc.2017.00139.

[249] G. Ashrafi, J. de Juan-Sanz, R.J. Farrell, T.A. Ryan, Molecular tuning of the axonal mitochondrial Ca^{2+} uniporter ensures metabolic flexibility of neurotransmission, Neuron. 105 (2020) 678-687.e5. https://doi.org/10.1016/j.neuron.2019.11.020.

[250] M. Patron, V. Granatiero, J. Espino, R. Rizzuto, D. De Stefani, MICU3 is a tissue-specific enhancer of mitochondrial calcium uptake, Cell Death Differ. 26 (2019) 179–195. https://doi.org/10.1038/s41418-018-0113-8.

[251] J.P. Lambert, T.S. Luongo, D. Tomar, P. Jadiya, E. Gao, X. Zhang, A.M. Lucchese, D.W. Kolmetzky, N.S. Shah, J.W. Elrod, MCUB regulates the molecular composition of the mitochondrial calcium uniporter channel to limit mitochondrial calcium overload during stress, Circulation. 140 (2019) 1720–1733. https://doi.org/10.1161/CIRCULATIONAHA.118.037968.

[252] R. Xu, M. Han, Y. Xu, X. Zhang, C. Zhang, D. Zhang, J. Ji, Y. Wei, S. Wang, B. Huang, A. Chen, Q. Zhang, W. Li, T. Sun, F. Wang, X. Li, J. Wang, Coiled-coil domain containing 109B is a HIF1α-regulated gene critical for progression of human gliomas, J. Transl. Med. 15 (2017) 165. https://doi.org/10.1186/s12967-017-1266-9.

[253] P.K. Chakraborty, S.B. Mustafi, X. Xiong, S.K.D. Dwivedi, V. Nesin, S. Saha, M. Zhang, D. Dhanasekaran, M. Jayaraman, R. Mannel, K. Moore, S. McMeekin, D. Yang, R. Zuna, K. Ding, L.

Tsiokas, R. Bhattacharya, P. Mukherjee, MICU1 drives glycolysis and chemoresistance in ovarian cancer, Nat. Commun. 8 (2017) 14634. https://doi.org/10.1038/ncomms14634.

[254] S. Patergnani, J.M. Suski, C. Agnoletto, A. Bononi, M. Bonora, E. De Marchi, C. Giorgi, S. Marchi, S. Missiroli, F. Poletti, A. Rimessi, J. Duszynski, M.R. Wieckowski, P. Pinton, Calcium signaling around Mitochondria Associated Membranes (MAMs), Cell Commun. Signal. 9 (2011) 19. https://doi.org/10.1186/1478-811X-9-19.

[255] C.T. Madreiter-Sokolowski, B. Gottschalk, W. Parichatikanond, E. Eroglu, C. Klec, M. Waldeck-Weiermair, R. Malli, W.F. Graier, Resveratrol specifically kills cancer cells by a devastating increase in the Ca^{2+} coupling between the greatly tethered endoplasmic reticulum and mitochondria, Cell. Physiol. Biochem. 39 (2016) 1404–1420. https://doi.org/10.1159/000447844.

[256] C.T. Madreiter-Sokolowski, B. Gottschalk, A.A. Sokolowski, R. Malli, W.F. Graier, Dynamic Control of mitochondrial Ca^{2+} levels as a survival strategy of cancer cells, Front. Cell Dev. Biol. 9 (2021) 614668. https://doi.org/10.3389/fcell.2021.614668.

[257] G.R. Bantug, M. Fischer, J. Grählert, M.L. Balmer, G. Unterstab, L. Develioglu, R. Steiner, L. Zhang, A.S.H. Costa, P.M. Gubser, A.V. Burgener, U. Sauder, J. Löliger, R. Belle, S. Dimeloe, J. Lötscher, A. Jauch, M. Recher, G. Hönger, M.N. Hall, P. Romero, C. Frezza, C. Hess, Mitochondria-endoplasmic reticulum contact sites function as immunometabolic hubs that orchestrate the rapid recall response of memory $CD8^+$ T Cells, Immunity. 48 (2018) 542-555.e6. https://doi.org/10.1016/j.immuni.2018.02.012.

[258] V.H. Sánchez-Vázquez, E. Martínez-Martínez, M.L. Gallegos-Gómez, J.M. Arias, G. Pallafacchina, R. Rizzuto, A. Guerrero-Hernández, Heterogeneity of the endoplasmic reticulum Ca^{2+} store determines colocalization with mitochondria, Cell Calcium. 109 (2023) 102688. https://doi.org/10.1016/j.ceca.2022.102688.

[259] J.G. Goetz, H. Genty, P. St.-Pierre, T. Dang, B. Joshi, R. Sauvé, W. Vogl, I.R. Nabi, Reversible interactions between smooth domains of the endoplasmic reticulum and mitochondria are regulated by physiological cytosolic Ca^{2+} levels, J. Cell Sci. 120 (2007) 3553–3564. https://doi.org/10.1242/jcs.03486.

[260] M. Kerkhofs, M. Bittremieux, G. Morciano, C. Giorgi, P. Pinton, J.B. Parys, G. Bultynck, Emerging molecular mechanisms in chemotherapy: Ca^{2+} signaling at the mitochondria-associated endoplasmic reticulum membranes, Cell Death Dis. 9 (2018) 334. https://doi.org/10.1038/s41419-017-0179-0.

[261] M. Doghman-Bouguerra, E. Lalli, ER-mitochondria interactions: Both strength and weakness within cancer cells, Biochim. Biophys. Acta - Mol. Cell Res. 1866 (2019) 650–662. https://doi.org/10.1016/j.bbamcr.2019.01.009.

[262] R. Wang, M. Wang, S. He, G. Sun, X. Sun, Targeting calcium homeostasis in myocardial ischemia/reperfusion injury: An overview of regulatory mechanisms and therapeutic reagents, Front. Pharmacol. 11 (2020) 872. https://doi.org/10.3389/fphar.2020.00872.

[263] K. Pittas, D.A. Vrachatis, C. Angelidis, S. Tsoucala, G. Giannopoulos, S. Deftereos, The role of calcium handling mechanisms in reperfusion injury, Curr. Pharm. Des. 24 (2018) 4077–4089. https://doi.org/10.2174/1381612825666181120155953.

[264] D. Garcia-Dorado, M. Ruiz-Meana, J. Inserte, A. Rodriguez-Sinovas, H.M. Piper, Calcium-mediated cell death during myocardial reperfusion, Cardiovasc. Res. 94 (2012) 168–180. https://doi.org/10.1093/cvr/cvs116.

[265] L. Guan, Z. Che, X. Meng, Y. Yu, M. Li, Z. Yu, H. Shi, D. Yang, M. Yu, MCU up-regulation contributes to myocardial ischemia-reperfusion Injury through calpain/OPA-1–mediated mitochondrial fusion/mitophagy Inhibition, J. Cell. Mol. Med. 23 (2019) 7830–7843. https://doi.org/10.1111/jcmm.14662.

[266] Q. Xue, H. Pei, Q. Liu, M. Zhao, J. Sun, E. Gao, X. Ma, L. Tao, MICU1 protects against myocardial ischemia/reperfusion injury and its control by the importer receptor Tom70, Cell Death Dis. 8 (2017) e2923. https://doi.org/10.1038/CDDIS.2017.280.

[267] J.Q. Kwong, X. Lu, R.N. Correll, J.A. Schwanekamp, R.J. Vagnozzi, M.A. Sargent, A.J. York, J. Zhang, D.M. Bers, J.D. Molkentin, The mitochondrial calcium uniporter selectively matches metabolic output to acute contractile stress in the heart, Cell Rep. 12 (2015) 15–22. https://doi.org/10.1016/j.celrep.2015.06.002.

[268] D.J. Hausenloy, E.A. Boston-Griffiths, D.M. Yellon, Cyclosporin A and cardioprotection: From investigative tool to therapeutic agent, Br. J. Pharmacol. 165 (2012) 1235–1245. https://doi.org/10.1111/j.1476-5381.2011.01700.x.

[269] K. Márta, P. Hasan, M. Rodríguez-Prados, M. Paillard, G. Hajnóczky, Pharmacological inhibition of the mitochondrial Ca^{2+} uniporter: Relevance for pathophysiology and human therapy, J. Mol. Cell. Cardiol. 151 (2021) 135–144. https://doi.org/10.1016/j.yjmcc.2020.09.014.

[270] J. Rieusset, J. Fauconnier, M. Paillard, E. Belaidi, E. Tubbs, M.A. Chauvin, A. Durand, A. Bravard, G. Teixeira, B. Bartosch, M. Michelet, P. Theurey, G. Vial, M. Demion, E. Blond, F.

Zoulim, L. Gomez, H. Vidal, A. Lacampagne, M. Ovize, Disruption of calcium transfer from ER to mitochondria links alterations of mitochondria-associated ER membrane integrity to hepatic insulin resistance, Diabetologia. 59 (2016) 614–623. https://doi.org/10.1007/s00125-015-3829-8.

[271] M. Paillard, E. Tubbs, P.A. Thiebaut, L. Gomez, J. Fauconnier, C.C. Da Silva, G. Teixeira, N. Mewton, E. Belaidi, A. Durand, M. Abrial, A. Lacampagne, J. Rieusset, M. Ovize, Depressing mitochondria-reticulum interactions protects cardiomyocytes from lethal hypoxia-reoxygenation injury, Circulation. 128 (2013) 1555–1565. https://doi.org/10.1161/CIRCULATIONAHA.113.001225.

[272] E. Tubbs, P. Theurey, G. Vial, N. Bendridi, A. Bravard, M.A. Chauvin, J. Ji-Cao, F. Zoulim, B. Bartosch, M. Ovize, H. Vidal, J. Rieusset, Mitochondria-associated endoplasmic reticulum membrane (MAM) integrity is required for insulin signaling and is implicated in hepatic insulin resistance, Diabetes. 63 (2014) 3279–3294. https://doi.org/10.2337/db13-1751.

[273] S. Han, F. Zhao, J. Hsia, X. Ma, Y. Liu, S. Torres, H. Fujioka, X. Zhu, The role of Mfn2 in the structure and function of endoplasmic reticulum-mitochondrial tethering in vivo, J. Cell Sci. 134 (2021) 253443. https://doi.org/10.1242/jcs.253443.

[274] L. Gomez, P.A. Thiebaut, M. Paillard, S. Ducreux, M. Abrial, C. Crola Da Silva, A. Durand, M.R. Alam, F. Van Coppenolle, S.S. Sheu, M. Ovize, The SR/ER-mitochondria calcium crosstalk is regulated by GSK3β during reperfusion injury, Cell Death Differ. 23 (2016) 313–322. https://doi.org/10.1038/cdd.2015.101.

[275] B. Bonneau, H. Ando, K. Kawaai, M. Hirose, H. Takahashi-Iwanaga, K. Mikoshiba, IRBIT controls apoptosis by interacting with the Bcl-2 homolog, Bcl2l10, and by promoting ER-mitochondria contact, Elife. 5 (2016) e19896. https://doi.org/10.7554/eLife.19896.

[276] C. Betz, D. Stracka, C. Prescianotto-Baschong, M. Frieden, N. Demaurex, M.N. Hall, mTOR complex 2-Akt signaling at mitochondria-associated endoplasmic reticulum membranes (MAM) regulates mitochondrial physiology, Proc. Natl. Acad. Sci. U. S. A. 110 (2013) 12526–12534. https://doi.org/10.1073/pnas.1302455110.

[277] M. Xue, T. Fang, H. Sun, Y. Cheng, T. Li, C. Xu, C. Tang, X. Liu, B. Sun, L. Chen, PACS-2 attenuates diabetic kidney disease via the enhancement of mitochondria-associated endoplasmic reticulum membrane formation, Cell Death Dis. 12 (2021) 1107. https://doi.org/10.1038/s41419-021-04408-x.

[278] X. Chen, L. Mi, G. Gu, X. Gao, M. Shi, Y. Chai, F. Chen, W. Yang, J. Zhang, Dysfunctional endoplasmic reticulum-mitochondrion coupling is associated with endoplasmic reticulum stress-induced apoptosis and neurological deficits in a rodent model of severe head injury, J. Neurotrauma. 39 (2022) 560–576. https://doi.org/10.1089/neu.2021.0347.

[279] L. Naia, P. Ly, S.I. Mota, C. Lopes, C. Maranga, P. Coelho, N. Gershoni-Emek, M. Ankarcrona, M. Geva, M.R. Hayden, A.C. Rego, The Sigma-1 receptor mediates pridopidine rescue of mitochondrial function in Huntington disease models, Neurotherapeutics. 18 (2021) 1017–1038. https://doi.org/10.1007/s13311-021-01022-9.

[280] N. Bernard-Marissal, J.J. Médard, H. Azzedine, R. Chrast, Dysfunction in endoplasmic reticulum-mitochondria crosstalk underlies SIGMAR1 loss of function mediated motor neuron degeneration, Brain. 138 (2015) 875–890. https://doi.org/10.1093/brain/awv008.

[281] H. Matsuzaki, T. Fujimoto, T. Ota, M. Ogawa, T. Tsunoda, K. Doi, M. Hamabashiri, M. Tanaka, S. Shirasawa, Tespa1 is a novel inositol 1,4,5-trisphosphate receptor binding protein in T and B lymphocytes, FEBS Open Bio. 2 (2012) 255–259. https://doi.org/10.1016/j.fob.2012.08.005.

[282] H. Matsuzaki, T. Fujimoto, M. Tanaka, S. Shirasawa, Tespa1 is a novel component of mitochondria-associated endoplasmic reticulum membranes and affects mitochondrial calcium flux, Biochem. Biophys. Res. Commun. 433 (2013) 322–326. https://doi.org/10.1016/j.bbrc.2013.02.099.