Biochimica et Biophysica Acta 1833 (2013) 3507–3517

Contents lists available at ScienceDirect

Biochimica et Biophysica Acta

journal homepage: www.elsevier.com/locate/bbamcr

Review

When ER stress reaches a dead end ☆

Hery Urra [a,b], Estefanie Dufey [a,b], Fernanda Lisbona [a,b], Diego Rojas-Rivera [a,b], Claudio Hetz [a,b,c,d,*]

[a] Institute of Biomedical Sciences, Center for Molecular Studies of the Cell, Santiago, Chile
[b] Biomedical Neuroscience Institute, Faculty of Medicine, University of Chile, Santiago, Chile
[c] Department of Immunology and Infectious Diseases, Harvard School of Public Health, Boston, MA 02115, USA
[d] Neurounion Biomedical Foundation, Santiago, Chile

ARTICLE INFO

Article history:
Received 30 April 2013
Received in revised form 25 July 2013
Accepted 30 July 2013
Available online 27 August 2013

Keywords:
Cell death
ER stress
Unfolded protein response
Adaptation
Apoptosis

ABSTRACT

Endoplasmic reticulum (ER) stress is a common feature of several physiological and pathological conditions affecting the function of the secretory pathway. To restore ER homeostasis, an orchestrated signaling pathway is engaged that is known as the unfolded protein response (UPR). The UPR has a primary function in stress adaptation and cell survival; however, under irreversible ER stress a switch to pro-apoptotic signaling events induces apoptosis of damaged cells. The mechanisms that initiate ER stress-dependent apoptosis are not fully understood. Several pathways have been described where we highlight the participation of the BCL-2 family of proteins and ER calcium release. In addition, recent findings also suggest that microRNAs and oxidative stress are relevant players on the transition from adaptive to cell death programs. Here we provide a global and integrated overview of the signaling networks that may determine the elimination of a cell under chronic ER stress. This article is part of a Special Section entitled: Cell Death Pathways. Guest Editors: Frank Madeo and Slaven Stekovic.

1. Introduction

The endoplasmic reticulum (ER) is a highly dynamic network composed by sac-like structures and tubules. The major function of the ER is the folding and maturation of nascent proteins that transit through the secretory pathway. Additionally, the ER is also implicated in bioenergetic processes such as lipid synthesis, in addition to calcium storage [1]. Different perturbations at the cellular level can affect ER homeostasis inducing the accumulation of unfolded proteins in the lumen of this organelle, a condition referred to as ER stress. ER stress activates a series of adaptive mechanisms known as the unfolded protein response (UPR) [2]. To alleviate ER stress, the UPR promotes an increased capacity of protein folding and clearance to reduce the amount of misfolded proteins at ER lumen [2,3]. However, under prolonged and irreversible ER stress, cells that become irreversibly damaged are eliminated by apoptosis [4,5].

ER stress-mediated cell death is executed by the canonical mitochondrial apoptosis pathway, where the BCL-2 family plays a crucial role [6]. Transcriptional and post-transcriptional mechanisms are activated to regulate pro-apoptotic members of the BCL-2 family that facilitate cytochrome c release from the mitochondria and calcium release from the ER to engage downstream apoptotic signaling events [7]. Recently, an explosion of novel mechanisms and components that can suppress or potentiate apoptosis during ER stress has been identified. In this review, we overview and integrate recent findings addressing the regulation of apoptosis under a condition of irreversible damage.

2. The unfolded protein response

Although the ER has been studied for decades, in the last ten years the impact of this organelle in cell physiology and disease had a major revival. One of the most studied functions of the ER is the folding and maturation of nascent proteins that are targeted to the plasma membrane, ER, lysosomes and Golgi apparatus or are secreted. Different conditions that interfere with ER function generate stress as a consequence of physiological demands or pathological perturbations. Independent of the nature of the stimuli that trigger ER stress, the UPR is initiated to restore ER homeostasis [2]. If the adaptive mechanisms of the UPR are not sufficient to recover homeostasis a switch to pro-apoptotic signals generates the death of irreversibly damaged cells. Therefore, a fine-tuning of pro-survival and pro-apoptotic mechanisms is necessary under physiological and pathological conditions to respond against ER stress [2,8].

ER stress is detected by at least three ER resident proteins: activating transcription factor 6 (ATF6), protein kinase RNA-like ER kinase (PERK) and inositol-requiring enzyme 1 (IRE1). These ER stress sensors induce different downstream signaling cascades that collectively constitute the UPR. ATF6 is synthetized as an inactive precursor coding for a bZIP transcription factor in the cytoplasmic domain. Under ER stress, ATF6 traffics to Golgi apparatus where S1P and S2P proteases cleave the cytosolic and transmembrane domains, releasing a fragment named ATF6f

☆ This article is part of a Special Section entitled: Cell Death Pathways. Guest Editors: Frank Madeo and Slaven Stekovic.

* Corresponding author at: Program of Cellular and Molecular Biology, Institute of Biomedical Sciences, University of Chile, P.O. Box 70086, Independencia 1027, Santiago, Chile. Tel.: +56 2 978 6506.
E-mail addresses: chetz@med.uchile.cl, chetz@hsph.harvard.edu (C. Hetz).

0167-4889/$ – see front matter © 2013 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.bbamcr.2013.07.024

[9]. ATF6f translocates to the nucleus regulating the transcription of genes involved in ER homeostasis, such as ER chaperones and ERAD components [10]. PERK is a transmembrane protein kinase that under ER stress conditions dimerizes and autophosphorylates, favoring the phosphorylation of eIF2α (eukaryotic translation initiation factor 2α). Phosphorylated eIF2α causes a global translational arrest as a fast adaptive reaction [11] and favors the selective translation of ATF4 (activating transcription factor 4) [12], regulating the expression genes involved in folding, oxidative stress and amino acid metabolism [13]. Finally, the IRE1 signaling branch is the most conserved arm of the UPR, and is present from yeast to mammals. There are two isoforms in mammals (IRE1α and IRE1β) that share two enzymatic activities at the cytoplasmic domain, kinase and endoribonuclease [14]. Under ER stress, IRE1α like PERK, dimerizes and autophosphorylates favoring a conformational change that activates the endoribonuclease domain. Active IRE1α catalyzes the unconventional splicing of the mRNA coding for XBP-1 (X box-binding protein 1) excising a 26-nucleotide intron, shifting the coding reading frame of this mRNA to generate an active transcription factor termed XBP-1s [15]. This transcription factor regulates the expression of genes related with folding, entry of proteins to the ER, ER-associated degradation (ERAD) and biogenesis of ER and Golgi, among others functions [16,17]. Although most of the signaling events downstream of UPR sensors favor adaptive responses, under chronic ER stress they signal toward induction of cell death. So far, the major death mechanism under ER stress is apoptosis [2], but it remains to be determined if other types of cell death, such as necrosis, necroptosis or deregulated autophagy, contribute to eliminate cells under irreversible ER stress. However, autophagy has been extensively described as a pro-survival mechanism to eliminate and degrade unfolded proteins and damaged organelles under ER stress [18,19]. Although several components have been described as responsible for the induction of apoptosis under ER stress [4,6,20], an integrative view is necessary to understand the checkpoints that control the switch from adaptive responses to pro-apoptotic phases.

3. Regulation of cell death under ER stress

3.1. PERK signaling and the regulation of ER stress-induced apoptosis

As mentioned above, under ER stress conditions PERK activates and phosphorylates eIF2α causing a translational arrest. This rapid response operates as an important pro-survival mechanism. Indeed, the deficiency of PERK or the expression of a non-phosphorylable form of eIF2α generates hypersensitivity to ER stress [11] or hypoxic conditions during tumor growth [21,22], supporting the idea of a physiological role of translational attenuation in supporting cell survival (Fig. 1). In addition to this rapid pro-survival mechanism, the PERK pathway can regulate a subset of genes related with autophagy, folding and redox metabolism through ATF4-mediated transcription decreasing the unfolded protein load [2].

Under prolonged ER stress, PERK signaling can trigger cell death. In fact, the artificial sustained activation of PERK, but not IRE1α, induces proliferation arrest and cell death in certain cellular models [23]. One major event responsible for this switch between pro-survival and pro-apoptotic responses is the induction of C/EBP homologous protein (CHOP: also named growth arrest and DNA-damage-inducible 153, GADD153), activated downstream of ATF4 [11].

The molecular mechanism that explains the pro-apoptotic role of CHOP involves in part the transcriptional regulation of several target genes. One of them is GADD34 (growth arrest and DNA damage inducible 34), a component of the GADD34–PP1 inducible eIF2α phosphatase [24]. GADD34 expression enhances reactive oxygen species (ROS) production possibly by promoting protein synthesis, which may trigger proteotoxicity [25]. Recently, it has been demonstrated that ATF4 and CHOP enhance the transcription of a subset of genes related to protein synthesis, but not apoptosis. This event induces cell death possibly due to augmented ROS production and ATP depletion [26], as suggested before [25]. In addition, the importance this process has been also elucidated by the generation of selective inhibitors for GADD34–PP1

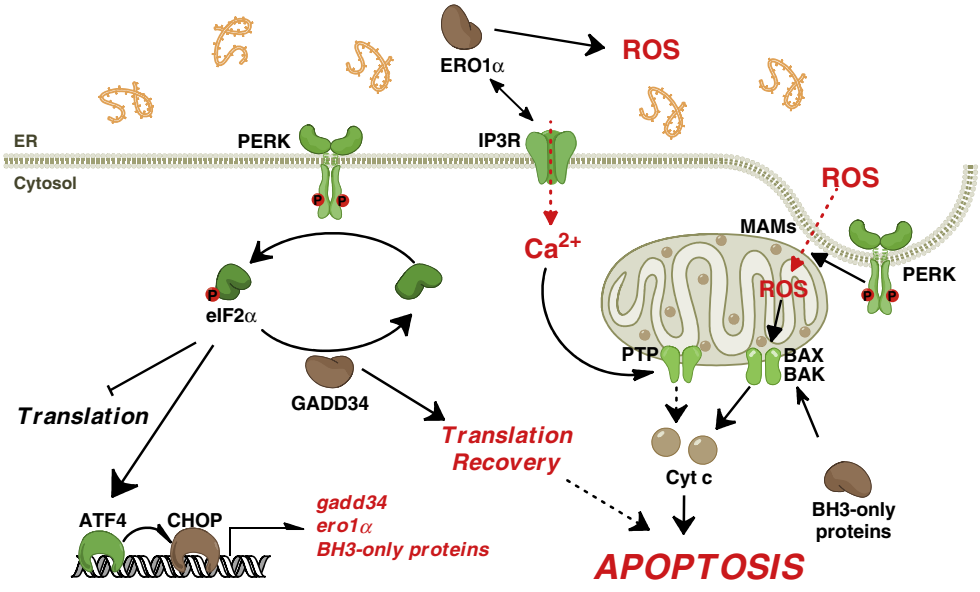

Fig. 1. PERK signaling in ER stress-induced apoptosis. Under ER stress, PERK activation induces eIF2α phosphorylation causing translational arrest. Phosphorylated eIF2α allows the selective translation of ATF4 that increases the levels of the transcription factor CHOP. CHOP induces the expression of several pro-apoptotic proteins such as GADD34, ERO1α and BH3-only proteins (BIM, PUMA and NOXA). Expression of GADD34 (a component of the GADD34-PP1 phosphatase) dephosphorylates eIF2α, restoring global translation of proteins that is involved in cytosolic ROS production and apoptosis. Increase of ERO1α at the ER lumen induces ROS in addition to the release of calcium by IPR3 regulation. Cytosolic calcium activates PTP triggering apoptosis. Moreover, the induction of BIM, PUMA and/or NOXA could activate BAX and BAK to release cytochrome c. In addition, PERK is required at the mitochondrial-associated ER membranes (MAMs) to modulate cytochrome c release and apoptosis by controlling calcium signaling and ROS production.

phosphatase activity that can decrease the protein synthesis rate and provide protection [27,28].

The most studied mechanism of cell death induced by CHOP is the regulation of the levels of several BCL-2 family members. Under ER stress, CHOP down-regulates the expression of BCL-2, sensitizing cells to apoptosis [29]. Additionally, the up-regulation of some pro-apoptotic components of the BCL-2 family, known as BH3-only proteins, is observed (see below). BIM levels are induced under ER stress conditions by a transcriptional mechanism involving CHOP [30]. Other reports also suggest an up-regulation of PUMA and NOXA [31], where CHOP and AP-1 seem to be responsible for their induction [32]. Importantly, the hypersensitivity of PERK-deficient cells to ER stress is rescued by silencing of NOXA, suggesting an important role of the BCL-2 family of proteins in the cell death process [33]. Recent studies demonstrate that BAX is also up-regulated during ER stress in cardiomyocyte models, showing that CHOP-deficient mice have less apoptotic cell death and lower caspase-3 activation related with a decrease of BAX levels [34].

As mentioned, another possible mechanism of action of CHOP is the modulation of the oxidative state of the ER. Overexpression of CHOP induces a depletion of glutathione and an exacerbated increase of ROS [29], possibly through the induction of ERO1α (ER oxidase 1α) [24]. ERO1α catalyzes the re-oxidation of several protein disulfide isomerases (PDIs) reconstituting their active state, a process responsible for the production of hydrogen peroxide (H_2O_2) at the ER lumen [35]. In addition, we recently reported that knocking down ATF4 increases H_2O_2 levels inside the ER and alters PDI expression [36]. The increases of ROS at the ER may sensitize cells to apoptosis under ER stress conditions [37]. Furthermore, this increase of cell death is abolished by CHOP depletion, reducing ERO1α levels, ROS production and apoptosis in pancreatic cells [38]. In addition, CHOP-induced ERO1α activates the inositol-1,4,5-trisphosphate receptor (IP_3R), leading to calcium release from ER [39]. This increase of cytoplasmic calcium may also contribute to apoptosis by affecting the opening of the mitochondrial permeability transition pore (PTP) [40]. Finally, PERK was recently shown to be located at the mitochondrial and ER contact sites, where it may modulate apoptosis by controlling local calcium release and ROS production [41].

Although a large amount of data suggests that PERK and CHOP are crucial components of the ER stress-induced cell death pathway, PERK or CHOP deficient cells still undergo apoptosis indicating the existence of other checkpoints and signaling events mediating this process. Furthermore, several reports in mouse models highlight the importance of CHOP and PERK in several diseases involving ER stress such as Parkinson's disease [42], diabetes [38], atherosclerosis [43], ALS [36], cardiac dysfunction [34] and liver damage induced by alcohol [44]. More studies are needed to understand the mechanisms explaining how PERK signaling can promote both cell survival and apoptosis depending on the intensity and duration of the stress stimuli.

3.2. Fine-tuning of IRE1α signaling in the regulation of ER stress-induced apoptosis

As discussed before, the IRE1α/XBP-1 pathway has an important pro-survival role through the up-regulation of a subset of target genes involved in protein folding and quality control. This adaptive response is evident in cells with artificial sustained activation of IRE1α [45]. In contrast with this pro-survival function, other reports showed that sustained IRE1α signaling could also engage apoptosis [46] (Fig. 2). IRE1α forms a complex with TRAF2 inducing the activation of ASK1/JNK, which triggers cell death [47,48]. Additionally, this pathway is also implicated in the induction of autophagy as a pro-survival mechanism [19,49]. Furthermore, IRE1α, in complex with TRAF2/IKK, activates NF-κB signaling that might lead to cell death [50].

In addition to the splicing of XBP-1, the endoribonuclease activity of IRE1α degrades a subset of mRNAs in a process known as regulated IRE1α-dependent decay (RIDD) [51]. Initially, RIDD was proposed as an adaptive mechanism of the UPR where certain mRNAs that encode for proteins of the secretory pathway that have a high tendency to misfold are degraded. However, additional evidence suggests that RIDD may also function as a pro-apoptotic mechanism by degrading mRNAs encoding for crucial proteins including ER chaperons such as BiP/Grp78 [51,52]. In addition using peptides derived from IRE1α that targets selectively the enzymatic activities of IRE1α revealed that

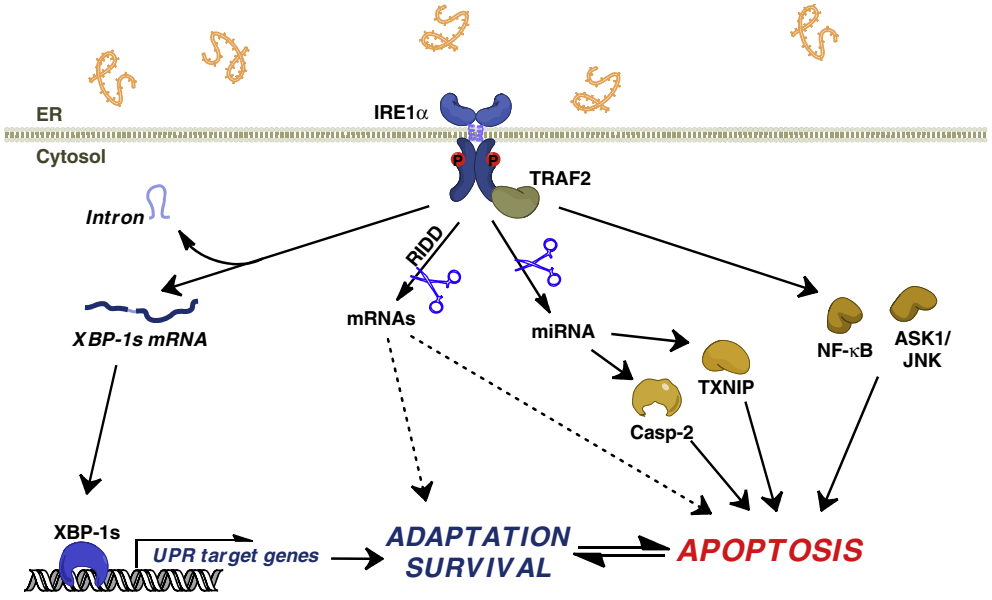

Fig. 2. Pleiotropic effects of IRE1α under ER stress. ER stress activates IRE1α inducing its dimerization and autophosphorylation, engaging the endoribonuclease domain. Active IRE1α processes the mRNA encoding for XBP-1, producing an active transcription factor termed XBP1s. XBP1s regulates an adaptive/survival response increasing the levels of proteins involved in protein folding and quality control. In addition, IRE1α degrades several mRNAs through a process known as RIDD. RIDD has a pro-survival function degrading mRNA coding for proteins with a high tendency to misfold and also has a pro-apoptotic activity by degrading mRNAs coding for key ER chaperones. In addition, the endoribonuclease activity of IRE1α can also cleave miRNAs that regulate the expression of pro-apoptotic proteins such as caspase-2 (casp-2) and TXNIP. The interaction of IRE1α with TRAF2 could also trigger the ASK1/JNK and NF-κB pathways promoting apoptosis under ER stress.

Table 1
Role of BCL-2 and TMBIM family members in adaptation and cell death under ER stress.

Member	Activation/regulation	Cell death phenotype	UPR signaling phenotype	References
BCL-2 family members				
BCL-2	↓ Transcription (CHOP)	Sensitive (KD)	Restores XBP-1 mRNA splicing in BIM/PUMA DKO cells. Interacts with IP_3R modulating the release of ER calcium.	[29,70,115]
BCL-X_L	No changes	Resistant (OE)	No changes in mRNA XBP-1 splicing. Interacts with IP_3R modulating the release of ER calcium.	[68,75,116]
MCL-1	↑ Transcription (IRE1α/ATF6)	Sensitive (KD)	No effects in mRNA XBP-1 splicing. Increases ER remodeling. Interacts and decreases the activity of BAX	[68,70–73]
BAX	↑ Transcription Translocation to the ER	Resistant (KO)	Interacts and modulates IRE1α signaling. Induces calcium release from the mitochondria and ER	[34,69,79,117,172]
BAK	↑ Transcription	Resistant (KO)	Interacts and modulates IRE1α signaling. Induces calcium release from the mitochondria and ER. Induces ER swelling regulated by BCL-X_L	[69,79,82,102,172]
BID	No changes in protein levels. Increase cleavage by caspase-2 (tBID)	Resistant (KO)	No effects	[80]
BIM	↑ Transcription (CHOP/Foxo3A). Translocation to the ER ↑ Protein stability	Resistant (KO)	Interacts and modulates IRE1α signaling. Potentiates BAX/BAX activation Promotes activation of caspase-12	[30,70,74,75]
PUMA	↑ Transcription (p53/CHOP/Foxo3A).	Resistant (KO)	Interacts and modulates IRE1α signaling Activates BAK and caspase-7	[31,32,70,74,76]
NOXA	↑ Transcription (p53/ATF4).	Resistant (KO)	No effects in IRE1α signaling. Activates BAK and caspase-7	[31,70,76,77]
BAD	↑ Activity by dephosphorylation	Unknown	Modulates IRE1α signaling	[70,78]
BMF	No changes	Unknown	Unknown	[79]
BOK	No changes	Sensitive (KO)	Induce ER/Golgi fragmentation Abnormal response to ER stress.	[79,83]
BNIP1	Unknown	Sensitive (OE)	Increases ER tubule fusion. Increases mitochondria fragmentation	[84,85]
BNIP3	↑ Transcription (Foxo3a)	Sensitive (OE)	Possible interaction with IRE1α. Increases eIF2α phosphorylation, CHOP and caspase-3 cleavage Increases mitochondrial calcium levels.	[70,86,87]
TMBIM family members				
TMBIM3/GRINA	↑ Transcription (PERK/ATF4)	Sensitive (KD and DKO TMBIM3/TMBIM6)	Interacts with IP_3R and modulates calcium homeostasis	[98]
TMBIM6/BI-1	No changes	Sensitive (KO)	Inhibits BAX-mediated apoptosis. Interacts and inhibits IRE1α signaling. Modulates ER calcium levels (IP_3R).	[139,145,146,150]

Cell death phenotype: Knockdown by siRNA or shRNA (KD), overexpression in cells (OE), knockout cells (KO) and double knockout cells (DKO).

attenuation of JNK signaling and RIDD could protect cells from ER stress-induced cell death [53]. However, on a recent report, the pharmacological inhibition of IRE1α endoribonuclease activity did not affect cell survival under acute ER stress [54]. Therefore, under ER stress the endoribonuclease activity of IRE1α may have opposite effects, where XBP-1 mRNA splicing promotes survival and RIDD in the long term could sensitize cells to apoptosis. This switch may depend on the duration and intensity of the stress stimuli. Finally, recent findings uncovered a new role of the endoribonuclease activity of IRE1α in the cleavage of microRNAs (miRNAs) and the regulation of the levels of caspase-2 [55] and TXNIP [56], having a significant impact on the activation of cell death programs (reviewed below).

Therefore, the selective activation of different enzymatic activities of IRE1α operates as signaling modules that generate divergent downstream outputs, reinforcing the idea that the fine-tuning of IRE1α could function as a stress integrator controlling the switch between adaptive and pro-apoptotic phases of the UPR [2,57,58]. This is a subject of current research that remains largely open for investigation.

3.3. ER stress and the BCL-2 family of proteins

Cytochrome c release from the mitochondria is a classical hallmark of ER stress-dependent apoptosis, associated with a collapse of mitochondrial transmembrane potential ($\Delta\Psi_m$) and an influx of calcium into the mitochondria. The permeabilization of the mitochondrial outer membrane (MOMP) is highly dependent on BAX and BAK, two central pro-apoptotic proteins of the BCL-2 family [40]. The BCL-2 family consists of ~20 members that share up to four conserved motifs known as BCL-2 homology domain (BH1-4) [59]. This family is subdivided into pro- and anti-apoptotic proteins that can be classified by the presence of one or more BH domains. The anti-apoptotic members of the family share all four BH domains (BCL-2, BCL-X_L, BCL-W and MCL-1) with the exception of BCL-2A1 and BCL-B. The pro-apoptotic proteins are divided into multidomain proteins that contain three BH domains (BAX and BAK) and BH3-only proteins that only have in common the BH3 domain (BID, BIK, BIM, PUMA NOXA and BAD) [59,60]. Additionally, another group of BH3-only proteins, termed BNIP, contains poor conservation in the BH domain (BNIP1, BNIP2 and BNIP3) [61,62]. The mechanism of action of anti-apoptotic members is the inhibition of the conformational activation of BAX and BAK through direct or indirect mechanisms [63]. Then, BH3-only proteins can be divided functionally into direct *activators* of BAX and BAK (i.e. BID, BIM and PUMA) or *sensitizers* that release pro-apoptotic activators sequestered by BCL-2 anti-apoptotic proteins (i.e. BAD and NOXA) [64].

The role of the BCL-2 family in adaptation and cell death under ER stress conditions has been widely reviewed [65–67]. Pro-apoptotic stimuli, like ER stress, induce the activation of certain BH3-only proteins by four different mechanisms: transcriptional regulation, posttranslational modifications, proteolytic cleavage and release from sequestering proteins (Table 1). BCL-2 family members have distinct locations in the cell, where anti-apoptotic members, such as BCL-2 and BCL-X_L, localize to the mitochondria and ER membrane, similar to BAK. On the other hand, BAX is mostly cytosolic but under apoptotic stimuli a C-terminal hydrophobic domain is exposed that allows the translocation to the

mitochondria and ER. Finally, BH3-only proteins are mostly cytosolic but interact with BCL-2 and BAX/BAK at different locations [67].

As mentioned, under ER stress CHOP down-regulates the expression of BCL-2 [29] but not BCL-X_L [68], sensitizing the cell to apoptosis. In addition, we have reported that BCL-2 family members such as BAK, BAX, BIM, and PUMA may also feedback to modulate the amplitude and kinetics of IRE1α signaling [69,70]. On the other hand, MCL-1 is up-regulated under ER stress through IRE1α and ATF6 signaling in melanoma, where this increase of protein levels contributes to cell survival [68,71,72]. Additionally, MCL-1 may not affect UPR signaling mediated by IRE1α [70], but influences ER membrane remodeling under ER stress induced by drugs that promote calcium depletion and ER stress [73].

Under ER stress, BIM levels are induced in different experimental systems by a transcriptional mechanism involving CHOP and FOXO3a [30,74]. In addition, BIM is regulated through dephosphorylation mediated by PP2A phosphatase blocking the proteasomal degradation of BIM [30]. BIM also translocates from microtubule fraction enriched in dynein to the ER inducing the activation of the ER-located caspase-12 [75]. PUMA levels are also up-regulated under ER stress conditions via CHOP, AP-1, p53 and FOXO3a transcription factors [31,32,74,76]. PUMA and NOXA [76] or BIM [70] deficient-cells are partially resistant to cell death induced by pharmacological inducers of ER stress. In addition, PUMA regulates BAK and caspase-7 activation [76] and modulates the amplitude of activation of IRE1α signaling [70]. NOXA is also up-regulated by p53 [76] and ATF4 [77], influencing cell death under ER stress. Unlike most of the BH3-only proteins, BAD is not regulated at the transcriptional level, instead BAD is activated by a rapid dephosphorylation increasing the rate of apoptosis under ER stress in neurons [78].

Other members of the BH3-only family, including BMF and BIK, are not regulated by ER stress at the transcriptional or post-transcriptional level [79] and their function in ER stress is unknown. In the case of BID, ER stress triggers caspase-2 activation, which then cleaves and activates BID (tBID) to promote apoptosis [80]. Unlike BAX, ER-located BAK has a unique function in the remodeling of ER structure [81], where it triggers swelling of the cisternae and vacuolization dependent on the co-expression of BCL-X_L [82]. Recently, it has been demonstrated that the multidomain pro-apoptotic component BOK locates to the ER and Golgi, and its expression contributes to apoptosis in cells treated with ER–Golgi trafficking inhibitor brefeldin A (an ER stressor) [83].

On the other hand, BNIP1 overexpression sensitizes cell to apoptosis affecting mitochondrial fragmentation and ER tubule fusion, dependent on the putative BH3 domain [84,85]. Under ER stress, BNIP3 is upregulated by the transcription factor FOXO3a through JNK-dependent signaling, increasing caspase-3 cleavage and increasing the levels of calcium at the mitochondria [86]. In addition, BNIP3 overexpression increases the phosphorylation of eIF2α and CHOP in mouse models of heart failure [86,87].

All data discussed in this section indicates an important role of several BCL-2 family members in the regulation of the UPR, calcium homeostasis, ER morphogenesis and cell death induced by ER stress. This concept has become also relevant in the context of several diseases where ER stress and protein misfolding have a relevant function [88]. For example, BIM deficiency protects against amyotrophic lateral sclerosis [89,90] and Huntington's disease [91]. PUMA deficiency has also protective effects in mouse models of ER stress-related diseases [92–94].

3.3.1. Calcium and the BCL-2 family of proteins

It is well described that depletion of ER calcium results in ER stress [95]. High ER calcium concentration, together with ER oxidizing environment, is essential for the proper folding of proteins. ER chaperones have high capacity and low affinity to calcium acting as calcium buffers [96,97]. More importantly, low levels of calcium decrease chaperone activity triggering ER stress. A few reports have shown that ER stress induced by tunicamycin induces the release of calcium from the ER through the IP_3R [39,98,99] or across the translocon [100] (Fig. 3). Both mechanisms involve chronic ER stress levels, and are associated with the opening of the PTP [101,102]. Besides, in an early stage of the ER stress response, an efflux of ER calcium to the mitochondria is observed that may promote an enhancement of mitochondrial metabolism [103,104]. Since the expression of several BCL-2 family members influences ER calcium release and steady state content (see examples in [102,105–114]), it is feasible to speculate that through this mechanism the BCL-2 family may influence the susceptibility of cells to ER stress (a folding effect), in addition to modulate the threshold to induce or not the release of cytochrome c from the mitochondria after chronic UPR activation. In fact, it has been shown that BCL-2 and BCL-X_L form a complex with the IP_3R, modulating the release of calcium from the ER and affecting cell death [115,116]. In addition, under ER stress BAX and BAK oligomerize at the ER membrane [117], correlating with ER calcium release to the cytosol and an increased sensitivity to ER stress [102]. Finally, although BAX and BAK double deficient cells are highly resistant to ER stress, we recently reported that the combination of ER stress agents with serum withdrawal triggers cytochrome c release and caspase-dependent apoptosis on a BAX and BAK-independent manner [118]. This pathway was also insensitive to the overexpression of BCL-X_L or the ablation of cyclophilin D expression, an essential component of the PTP [119]. These results suggest the existence of an alternative pore for the release of cytochrome c under ER stress.

3.3.2. Caspase activation and ER stress

A hallmark of apoptosis is the activation of caspases where the BCL-2 family has a crucial role regulating their engagement under ER stress [120]. A pioneering study revealed the expression of an ER-located caspase in murine cells known as caspase-12 [121], as described latter for caspase-4 in human cells [122,123]. The molecular mechanism underlying caspases-12 activation is not well understood and depending on the experimental system may involve BIM translocation to the ER [75], BAX and BAK activation at the ER [114], the activation of IRE1α and the recruitment of TRAF2 [124] and calpain activation [125–127], among other mechanisms. However, although caspase-12 processing is a well-demonstrated marker of ER stress, its contribution to cell death has been questioned. Caspase-12 is closer to pro-inflammatory caspases and may influence susceptibility to pathogens [128–131]. All this mechanisms have in common the release of cytochrome c downstream activation of caspase-9. Caspase-8 is also activated promoting cytochrome c release via cleavage of BID [132]. Furthermore, caspase-4 can directly cleave pro-caspase-9 to trigger apoptosis under ER stress in neurons [133] and neutrophils [134].

As mentioned above, the cleavage of BID by caspase-2 contributes to the induction of apoptosis by sustained ER stress [80]. Furthermore, caspase-2 has been localized at the ER where it is activated triggering apoptosis depending on executer caspases -3 and -7 [135]. In addition, a similar mechanism was reported in cells treated with the proteasome inhibitor bortezomib [136], an anticancer drug that induces ER stress [137]. Interestingly, a new mechanism has been described that might promote cell death up-regulating caspase-2 levels under ER stress by the direct cleavage of specific microRNAs [55] (reviewed below). In summary, data relating caspase activation to ER stress mediated cell death is available but with little connection between the different studies. Most of the research had focused on understanding upstream signaling events involved in cell death under ER stress.

3.4. The TMBIM family of proteins and ER stress

A new family of highly conserved cell death regulators is emerging in recent years known as the BAX inhibitor 1 (BI-1) or transmembrane BAX inhibitor-containing motif (TMBIM) family [138]. This group of genes is present in humans, insects, plants, yeasts and viruses, among other species [139,140]. A common feature of this family is the presence

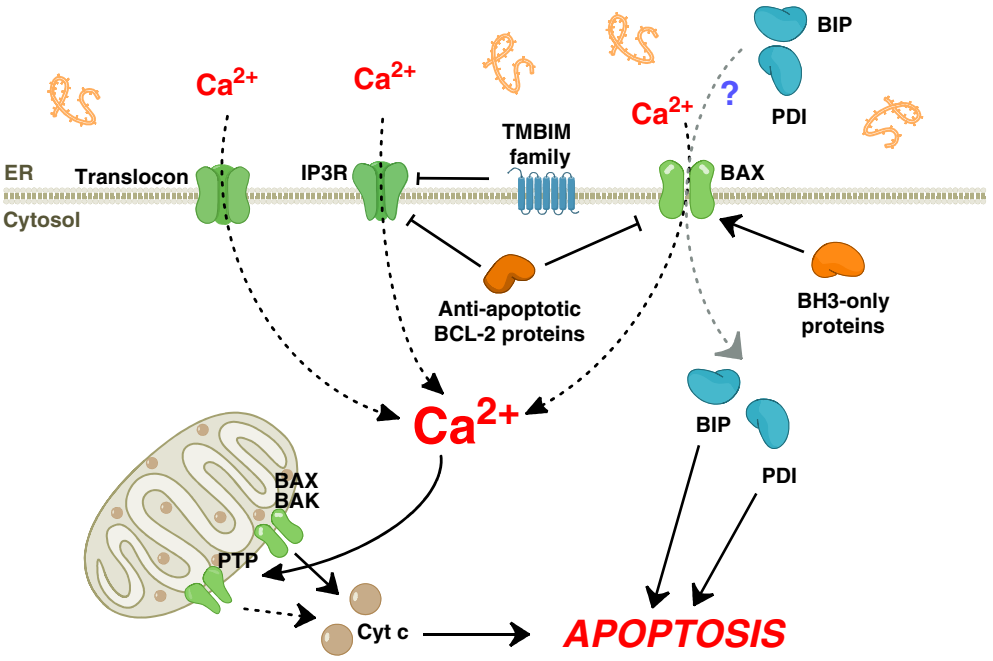

Fig. 3. Calcium regulation and ER permeabilization during chronic ER stress. During ER stress, cytosolic calcium increases due to activation of the IP_3R and possibly the ER translocon. IP_3R activity is tightly regulated by a physical interaction with anti-apoptotic BCL-2 proteins (BCL-2 and BCL-X_L) and TMBIM family members (BI-1/TMBIM6 and GRINA/TMBIM3). Increase of cytosolic calcium induces apoptosis by affecting BAX/BAX oligomerization (MOMP) and also PTP at the mitochondria. In addition, ER stress affects ER permeability, releasing ER foldases such as BiP and PDIs to the cytosol possibly mediated by BAX/BAK. BiP at the cytosol translocates to the plasma membrane, where it serves as a cell surface receptor for the pro-apoptotic protein Par-4. In addition, PDIs translocate to ER–mitochondria contact sites, inducing MOMP and apoptosis.

of multiple hydrophobic transmembrane domains (6 to 7) which defines through bioinformatics analysis a consensus domain at the C-terminal region of the proteins named UPF0005 [141].

The TMBIM family is composed of at least 6 members with different localizations in membranous compartments. The TMBIM family includes BI-1/TMBIM6 (BAX inhibitor 1), GRINA/TMBIM3 (glutamate receptor, ionotropic, N-methyl-D-asparate associated protein 1), GAAP/TMBIM4 (Golgi anti-apoptotic protein) and LFG/TMBIM2 (Lifeguard), which are all located to the ER membrane. In addition, LFG/TMBIM2 is also located at the plasma membrane, whereas GAAP/TMBIM4 is expressed to the Golgi apparatus. GHITM/TMBIM5 (Growth-hormone inducible transmembrane protein) is expressed at the mitochondrial membrane and RECS1/TMBIM1 (Responsive to centrifugal force and shear stress gene 1) is located to acidic vesicles, the Golgi apparatus and plasma membrane [138,142–144].

In general, all TMBIM family members have demonstrated anti-apoptotic activities, where only GRINA/TMBIM3 and BI-1/TMBIM6 are reported to be involved in ER stress-mediated cell death (Table 1) [98,145,146]. Interestingly, the BI-1/TMBIM6 homologue *Ybh3p* in yeast was shown to induce cell death dependent on a putative BH3 domain [147], whereas other reports indicated pro-survival effects under ER stress in yeast [131,148]. BI-1/TMBIM6 is highly conserved and is able to suppress apoptotic cell death mediated by BAX overexpression [149]. BI-1/TMBIM6 overexpression protects cells against apoptosis induced by nutrient deprivation, DNA damage and oxidative stress, but not from extrinsic cell death stimulation [149]. Similar findings were reported for RECS1/TMBIM1 and LFG/TMBIM2 [138,142]. BI-1/TMBIM6 deficient cells are highly susceptible to ER stress [139]. At the molecular level it has been shown that BI-1/TMBIM6 increases ER calcium content, affecting stimulated calcium release [139,150,151].

Several indirect studies suggest a functional connection between BCL-2 and TMBIM family of proteins. For example, co-immunoprecipitation experiments suggest that BI-1/TMBIM6 interacts with BCL-2 and BCL-X_L, but not with BAX [138,152]. Studies in BI-1/TMBIM6-deficient mice revealed that this protein is an important regulator of apoptosis under conditions of ER stress *in vivo* [139,145]. Our group demonstrated that BI-1/TMBIM6-deficient cells show IRE1α hyper-activation associated with increased levels of XBP-1s and its downstream transcriptional targets [146]. The same pathway can modulate the activation of autophagy [49]. The inhibitory effects of BI-1/TMBIM6 in the UPR were associated with the formation of a stable complex with the cytosolic domain of IRE1α. *In vivo* experiments in BI-1/TMBIM6-deficient mice or *Drosophila melanogaster* overexpressing BI-1/TMBIM6, indicate a suppression of IRE1α activity under experimental ER stress [146]. Similar results were provided on a brain injury model [153] and diabetes [154].

We recently reported that GRINA/TMBIM3 has an important activity controlling cell death induced by ER stress. GRINA/TMBIM3 expression is increased in both cellular and animal models of ER stress, mostly controlled by the PERK/ATF4 pathway [98]. In addition, GRINA/TMBIM3 is able to modulate calcium homeostasis of the ER through a physical association with the IP_3R (Fig. 3). BI-1/TMBIM6 and GRINA/TMBIM3 have a synergistic effect on calcium regulation in cellular models, associated with the formation of a protein complex between both proteins [98]. Similarly, double deficient flies for these two genes showed synergistic effects on the control of cell death *in vivo* on an ER stress model. Remarkably, the manipulation of GRINA/TMBIM3 levels in zebrafish embryos revealed an essential role in the control of apoptosis during neural development and experimental models of ER stress [98]. Interestingly, GAAP/TMBIM4 [155] is also able to control the ER calcium concentration possibly downstream of the BCL-2 family where they reduce ER calcium content [150]. Together, these reports indicate a functional interconnection between TMBIM family members and their role in the control of cell death and ER calcium homeostasis. More studies are needed to address the possible functional interconnection between the TMBIM and BCL-2 family of proteins.

3.5. ER permeabilization during ER stress: a novel mechanism in cell death?

Only few studies have suggested that ER membrane permeability to luminal proteins is altered in cells undergoing apoptosis, in addition to

modulating the three-dimensional structure of the ER [79,81]. For example, a recent report indicates that the regulation of ER membrane permeability by BCL-2 family members could operate as an important molecular mechanism of ER stress-induced apoptosis. It has been demonstrated that ER luminal proteins are released into the cytosol during ER stress-induced apoptosis in a BAK/BAX-dependent manner (Fig. 3) [156]. Furthermore, recombinant BAX activated by t-BID triggers ER membrane permeability *in vitro* [157]. In addition, BAX/BAK-dependent ER permeabilization is antagonized by BCL-X_L and enhanced by BH3-only proteins [156]. However, the functional role of the ER permeabilization is unknown.

Interestingly, BiP and PDI are released into the cytosol during ER stress [156]. Although, ER-located chaperons have pro-survival activities, the mislocation of these foldases into the cytosol may have pro-apoptotic activities. In cells undergoing ER stress, BiP is translocated to the plasma membrane, where it may act as a cell surface receptor for the pro-apoptotic protein Par-4 to activate FADD/caspases-8/caspases-3 apoptotic signaling pathway [158]. Importantly, PDI has a pro-apoptotic activity through its translocation to ER–mitochondria contact sites where it induces MOMP [159]. Although these reports suggest a role of ER permeabilization in ER stress, most of the data available is correlative; therefore, more studies are needed to define the actual impact of ER permeabilization to ER stress-mediated cell death.

3.6. miRNA and ER stress: orchestrating apoptotic cell death

In the last few years, it is becoming evident that miRNAs have a profound impact in many pathological and physiological processes, affecting cell proliferation, apoptosis and stress responses. miRNAs are non-coding single-stranded RNAs of ~22 nucleotides which are derived from endogenously produced pre-miRNAs (precursors) of 75–80 nucleotides containing a hairpin (stem-loop) structure. miRNAs regulate their target genes by binding to the 3′ UTR region of the mRNA with consequent transcriptional or translational repression [160].

Only a few recent studies have related ER stress-induced cell death with miRNA biology (Fig. 4) [161,162]. For example, *miR-708* is controlled by ER stress signaling through CHOP expression. Additionally, *miR-708* targets rhodopsin mRNA repressing its protein expression [163], highlighting a possible role of *miR-708* in the modulation of the expression of proteins with a high tendency to misfold like rhodopsin [163]. As mentioned above, the XBP-1 pathway is an important pro-survival response; therefore, the modulation of its levels might affect cell fate under ER stress. In fact, the PERK pathway induces the *miR-30c-2**, which specifically represses XBP-1 expression during ER stress [164], like *miR-214* [165]. In addition, the reduced levels of *miR-214*, *miR-199a-3p* and *miR-199a-5p* in hepatocellular carcinoma impact apoptosis levels [165]. A recent report indicates that *miR-199a-5p* directly represses the expression of IRE1α, ATF6 and Grp78, promoting the survival of hepatocytes under sustained ER stress [166], contrasting with a previous report [165].

As mentioned, a recent study indicates that during ER stress IRE1α cleaves certain miRNAs. Normally, the translation of caspase-2 mRNA is repressed by *miR-17~34a~96~125b*; however, sustained activation of IRE1α directly reduces the levels of these miRNAs, increasing the levels of caspase-2 mRNA translation [55]. In addition, it has been described that hyperactivated IRE1α stabilizes the mRNA encoding thioredoxin-interacting protein (TXNIP) by reducing its inhibitory miRNA, *miR-17*. TXNIP protein in turn activates the NLRP3 inflammasome to cleave procaspase-1 to its active form, thereby causing maturation and secretion of the inflammatory cytokine IL-1β and programmed cell death of pancreatic beta cells [56]. Furthermore, a recent study suggested that *miR-1291* represses the expression of IRE1α through a specific site at the 5′ UTR of IRE1α mRNA [167], which may impact the activity of IRE1α in apoptosis.

Furthermore, it was demonstrated that miRNAs belonging to the *miR-106b-25* cluster have an important role in ER stress-induced apoptosis through the regulation of pro-apoptotic BCL-2 family members [168]. Under ER stress the *miR-106b-25* cluster is repressed by ATF4, contributing to the increase of BIM [168]. Other studies have shown that the up-regulation of the *miR-23a~27a~24-2* cluster induces apoptosis, increasing the levels of pro-apoptotic components (i.e., CHOP and TRIB3) [169]. PERK induces *miR-211* through ATF4 induction, which in turn attenuates stress-dependent expression of CHOP, preventing its premature accumulation [170]. Finally, the deficiency of *DICER*, a crucial component of the miRNA machinery, leads to exacerbated sensitivity to ER stress [166]. In summary, modulation of miRNA biology is emerging as a new layer of complexity on the regulation of ER stress-mediated cell death.

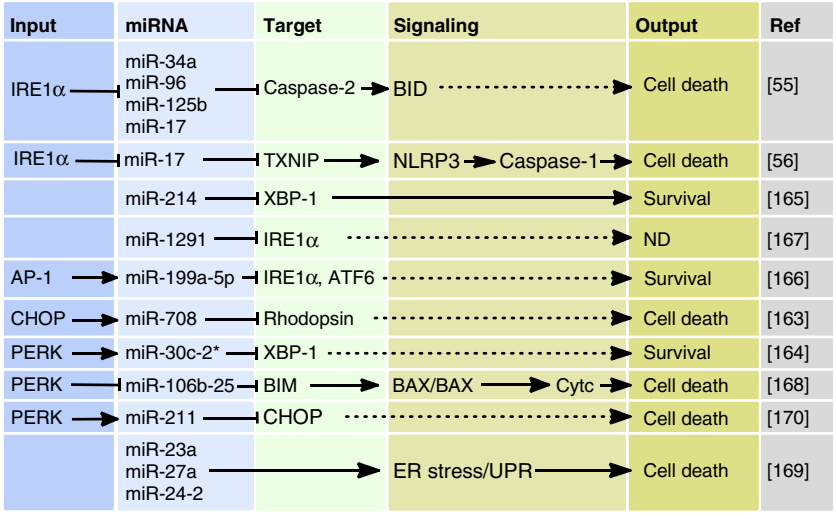

Fig. 4. miRNAs and ER stress-induced apoptosis. Caspase-2 expression is negatively regulated by *miR-17~34a~96~125b*. Under ER stress, IRE1α cleaves specifically these miRNAs increasing the levels of caspase-2, cleavage of BID and apoptosis. In addition, the cleavage of *miR-17* promotes the translation of TXNIP activating the NLRP3 inflammasome and apoptosis influenced by caspase-1. In addition, *miR-1291* can regulate negatively the levels of IRE1α. XBP-1 levels are regulated by *miR-214* and *miR-30c2** downstream of PERK activation, sensitizing cells to apoptosis. CHOP induces the expression of *miR-708* regulating the levels of rhodopsin. In addition, under ER stress the *miR-106b-25* cluster is repressed by ATF4 and NRF2, increasing the levels of BIM and apoptosis. In addition, PERK induces *miR-211* through eIF2α phosphorylation and ATF4 induction, which in turn attenuates stress-dependent expression of CHOP. The *miR-23a~27a~24-2* cluster induces apoptosis by ER stress modulating CHOP and TRIB3 (ND: non-determined).

4. Concluding remarks

The UPR is constituted by several signaling pathways that regulate both stress adaptation and cell death. As discussed here, different molecular mechanisms participate in ER stress-induced apoptosis; however, the molecular switches mediating the transition from adaptive to pro-apoptotic responses are not fully understood. So far, the most accepted components mediating cell death under ER stress are downstream of PERK, including the up-regulation of CHOP, which induces translation recovery, ROS production, BCL-2 family expression (i.e. BIM and PUMA), and ER calcium release. Despite of all this evidence, the final outcome of PERK deficient cells under ER stress is decreased survival [171], suggesting that the regulation of cell death by ER stress is highly complex and may depend in an additional crosstalk with other signaling pathways. IRE1α also has pleiotropic effects, controlling both pro- and anti-apoptotic events including RIDD, miRNA cleavage and JNK activation, in addition to XBP-1 mRNA splicing.

The intersection of ER stress signaling events may involve multiple simultaneous steps at the level of UPR stress sensor regulation, interactions between BCL-2/TMBIM family members and calcium release. In addition, it is important to highlight that data derived from the use of knockout cells for UPR components indicate that none of the single components of the pathway are absolutely necessary for the induction of apoptosis under ER stress. This suggests that the UPR controls cell death by a global network of events that are interconnected and highly regulated (Fig. 5).

Here, we have overviewed distinct molecular mechanisms that may induce or sensitize cells to apoptosis; however, the switching mechanism that turns off the adaptive UPR responses and engages pro-apoptotic signaling is still missing. It is interesting to note that several apoptosis regulators physically interact with IRE1α at the ER membrane, determining the kinetic and amplitude of its signaling through the formation of a platform known as the UPRosome [2,5]. We speculate that this mechanism may allow BCL-2 family members to operate as “stress sentinels” that may then contribute to switching IRE1α signaling from adaptive to pro-apoptotic outputs. Identifying the components involved in ER stress-mediated cell death may lead to the identification of novel targets for disease intervention based on the fact that ER stress is implicated on a variety of human diseases ranging from cancer, diabetes, neurodegeneration and inflammatory diseases. The specific networks that govern ER stress-induced apoptosis may depend on the nature of the stimuli, their intensity and even the cell type affected. The scenario in full organs may be even more complex where several independent pathological inputs could converge and synergize into the activation of UPR-dependent cell death. It is evident from overviewing the field that the mechanisms of apoptosis under ER stress are poorly understood and a broad and integrative approach is needed to uncover the actual mechanisms governing this essential process.

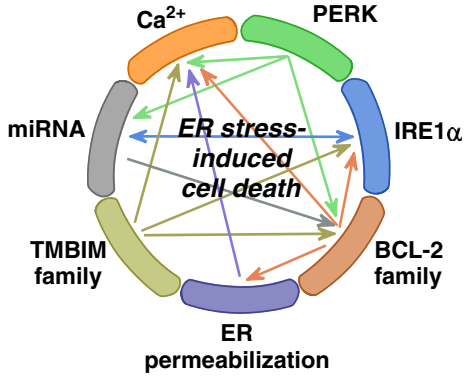

Fig. 5. Crosslink of pro-apoptotic pathways under ER stress. An overview of the pathways involved in apoptosis during ER stress are summarized depicting a high degree of cross talk and integration.

Acknowledgements

We thank Ute Woehlbier for her insightful comments. This work was funded by FONDEF D11I1007, Ring Initiative ACT1109, Millennium Institute No. P09-015-F, the Alzheimer's Association, FONDECYT no. 1100176, the Muscular Dystrophy Association, ALS Therapy Alliance (CH), FONDECYT #3130365 post-doctoral grant (DRR) and CONICYT PhD fellowship (HU, ED, FL).

References

[1] M.J. Berridge, The endoplasmic reticulum: a multifunctional signaling organelle, Cell Calcium 32 (2002) 235–249.
[2] C. Hetz, The unfolded protein response: controlling cell fate decisions under ER stress and beyond, Nat. Rev. Mol. Cell Biol. 13 (2012) 89–102.
[3] P. Walter, D. Ron, The unfolded protein response: from stress pathway to homeostatic regulation, Science 334 (2011) 1081–1086.
[4] I. Tabas, D. Ron, Integrating the mechanisms of apoptosis induced by endoplasmic reticulum stress, Nat. Cell Biol. 13 (2011) 184–190.
[5] U. Woehlbier, C. Hetz, Modulating stress responses by the UPRosome: a matter of life and death, Trends Biochem. Sci. 36 (2011) 329–337.
[6] G.C. Shore, F.R. Papa, S.A. Oakes, Signaling cell death from the endoplasmic reticulum stress response, Curr. Opin. Cell Biol. 23 (2011) 143–149.
[7] D. Rodriguez, D. Rojas-Rivera, C. Hetz, Integrating stress signals at the endoplasmic reticulum: the BCL-2 protein family rheostat, Biochim. Biophys. Acta 1813 (2011) 564–574.
[8] D. Ron, P. Walter, Signal integration in the endoplasmic reticulum unfolded protein response, Nat. Rev. Mol. Cell Biol. 8 (2007) 519–529.
[9] K. Haze, H. Yoshida, H. Yanagi, T. Yura, K. Mori, Mammalian transcription factor ATF6 is synthesized as a transmembrane protein and activated by proteolysis in response to endoplasmic reticulum stress, Mol. Biol. Cell 10 (1999) 3787–3799.
[10] R. Asada, S. Kanemoto, S. Kondo, A. Saito, K. Imaizumi, The signalling from endoplasmic reticulum-resident bZIP transcription factors involved in diverse cellular physiology, J. Biochem. 149 (2011) 507–518.
[11] H.P. Harding, I. Novoa, Y. Zhang, H. Zeng, R. Wek, M. Schapira, D. Ron, Regulated translation initiation controls stress-induced gene expression in mammalian cells, Mol. Cell 6 (2000) 1099–1108.
[12] K.M. Vattem, R.C. Wek, Reinitiation involving upstream ORFs regulates ATF4 mRNA translation in mammalian cells, Proc. Natl. Acad. Sci. U. S. A. 101 (2004) 11269–11274.
[13] H.P. Harding, Y. Zhang, H. Zeng, I. Novoa, P.D. Lu, M. Calfon, N. Sadri, C. Yun, B. Popko, R. Paules, D.F. Stojdl, J.C. Bell, T. Hettmann, J.M. Leiden, D. Ron, An integrated stress response regulates amino acid metabolism and resistance to oxidative stress, Mol. Cell 11 (2003) 619–633.
[14] T.N. Gonzalez, P. Walter, Ire1p: a kinase and site-specific endoribonuclease, Methods Mol. Biol. 160 (2001) 25–36.
[15] M. Calfon, H. Zeng, F. Urano, J.H. Till, S.R. Hubbard, H.P. Harding, S.G. Clark, D. Ron, IRE1 couples endoplasmic reticulum load to secretory capacity by processing the XBP-1 mRNA, Nature 415 (2002) 92–96.
[16] D. Acosta-Alvear, Y. Zhou, A. Blais, M. Tsikitis, N.H. Lents, C. Arias, C.J. Lennon, Y. Kluger, B.D. Dynlacht, XBP1 controls diverse cell type- and condition-specific transcriptional regulatory networks, Mol. Cell 27 (2007) 53–66.
[17] A.H. Lee, N.N. Iwakoshi, L.H. Glimcher, XBP-1 regulates a subset of endoplasmic reticulum resident chaperone genes in the unfolded protein response, Mol. Cell. Biol. 23 (2003) 7448–7459.
[18] G. Kroemer, G. Marino, B. Levine, Autophagy and the integrated stress response, Mol. Cell 40 (2010) 280–293.
[19] M. Ogata, S. Hino, A. Saito, K. Morikawa, S. Kondo, S. Kanemoto, T. Murakami, M. Taniguchi, I. Tanii, K. Yoshinaga, S. Shiosaka, J.A. Hammarback, F. Urano, K. Imaizumi, Autophagy is activated for cell survival after endoplasmic reticulum stress, Mol. Cell. Biol. 26 (2006) 9220–9231.
[20] A.M. Gorman, S.J. Healy, R. Jager, A. Samali, Stress management at the ER: regulators of ER stress-induced apoptosis, Pharmacol. Ther. 134 (2012) 306–316.
[21] M. Bi, C. Naczki, M. Koritzinsky, D. Fels, J. Blais, N. Hu, H. Harding, I. Novoa, M. Varia, J. Raleigh, D. Scheuner, R.J. Kaufman, J. Bell, D. Ron, B.G. Wouters, C. Koumenis, ER stress-regulated translation increases tolerance to extreme hypoxia and promotes tumor growth, EMBO J. 24 (2005) 3470–3481.
[22] C. Koumenis, C. Naczki, M. Koritzinsky, S. Rastani, A. Diehl, N. Sonenberg, A. Koromilas, B.G. Wouters, Regulation of protein synthesis by hypoxia via activation of the endoplasmic reticulum kinase PERK and phosphorylation of the translation initiation factor eIF2alpha, Mol. Cell. Biol. 22 (2002) 7405–7416.
[23] J.H. Lin, H. Li, Y. Zhang, D. Ron, P. Walter, Divergent effects of PERK and IRE1 signaling on cell viability, PLoS One 4 (2009) e4170.
[24] S.J. Marciniak, C.Y. Yun, S. Oyadomari, I. Novoa, Y. Zhang, R. Jungreis, K. Nagata, H.P. Harding, D. Ron, CHOP induces death by promoting protein synthesis and oxidation in the stressed endoplasmic reticulum, Genes Dev. 18 (2004) 3066–3077.
[25] E. Kojima, A. Takeuchi, M. Haneda, A. Yagi, T. Hasegawa, K. Yamaki, K. Takeda, S. Akira, K. Shimokata, K. Isobe, The function of GADD34 is a recovery from a shutoff of protein synthesis induced by ER stress: elucidation by GADD34-deficient mice, FASEB J. 17 (2003) 1573–1575.
[26] J. Han, S.H. Back, J. Hur, Y.H. Lin, R. Gildersleeve, J. Shan, C.L. Yuan, D. Krokowski, S. Wang, M. Hatzoglou, M.S. Kilberg, M.A. Sartor, R.J. Kaufman, ER-stress-induced

transcriptional regulation increases protein synthesis leading to cell death, Nat. Cell Biol. 15 (2013) 481–490.
[27] M. Boyce, K.F. Bryant, C. Jousse, K. Long, H.P. Harding, D. Scheuner, R.J. Kaufman, D. Ma, D.M. Coen, D. Ron, J. Yuan, A selective inhibitor of eIF2alpha dephosphorylation protects cells from ER stress, Science 307 (2005) 935–939.
[28] P. Tsaytler, H.P. Harding, D. Ron, A. Bertolotti, Selective inhibition of a regulatory subunit of protein phosphatase 1 restores proteostasis, Science 332 (2011) 91–94.
[29] K.D. McCullough, J.L. Martindale, L.O. Klotz, T.Y. Aw, N.J. Holbrook, Gadd153 sensitizes cells to endoplasmic reticulum stress by down-regulating Bcl2 and perturbing the cellular redox state, Mol. Cell. Biol. 21 (2001) 1249–1259.
[30] H. Puthalakath, L.A. O'Reilly, P. Gunn, L. Lee, P.N. Kelly, N.D. Huntington, P.D. Hughes, E.M. Michalak, J. McKimm-Breschkin, N. Motoyama, T. Gotoh, S. Akira, P. Bouillet, A. Strasser, ER stress triggers apoptosis by activating BH3-only protein Bim, Cell 129 (2007) 1337–1349.
[31] C. Reimertz, D. Kogel, A. Rami, T. Chittenden, J.H. Prehn, Gene expression during ER stress-induced apoptosis in neurons: induction of the BH3-only protein Bbc3/PUMA and activation of the mitochondrial apoptosis pathway, J. Cell Biol. 162 (2003) 587–597.
[32] S.C. Cazanave, N.A. Elmi, Y. Akazawa, S.F. Bronk, J.L. Mott, G.J. Gores, CHOP and AP-1 cooperatively mediate PUMA expression during lipoapoptosis, Am. J. Physiol. Gastrointest. Liver Physiol. 299 (2010) G236–G243.
[33] S. Gupta, Z. Giricz, A. Natoni, N. Donnelly, S. Deegan, E. Szegezdi, A. Samali, NOXA contributes to the sensitivity of PERK-deficient cells to ER stress, FEBS Lett. 586 (2012) 4023–4030.
[34] H.Y. Fu, K. Okada, Y. Liao, O. Tsukamoto, T. Isomura, M. Asai, T. Sawada, K. Okuda, Y. Asano, S. Sanada, H. Asanuma, M. Asakura, S. Takashima, I. Komuro, M. Kitakaze, T. Minamino, Ablation of C/EBP homologous protein attenuates endoplasmic reticulum-mediated apoptosis and cardiac dysfunction induced by pressure overload, Circulation 122 (2010) 361–369.
[35] T.J. Tavender, N.J. Bulleid, Molecular mechanisms regulating oxidative activity of the Ero1 family in the endoplasmic reticulum, Antioxid. Redox Signal. 13 (2010) 1177–1187.
[36] S. Matus, E. Lopez, V. Valenzuela, M. Nasiff, C. Hetz, Functional role of the transcription factor ATF4 in the pathogenesis of amyotrophic lateral sclerosis, PLoS One 8 (2013) e66672.
[37] J.D. Malhotra, R.J. Kaufman, Endoplasmic reticulum stress and oxidative stress: a vicious cycle or a double-edged sword? Antioxid. Redox Signal. 9 (2007) 2277–2293.
[38] B. Song, D. Scheuner, D. Ron, S. Pennathur, R.J. Kaufman, Chop deletion reduces oxidative stress, improves beta cell function, and promotes cell survival in multiple mouse models of diabetes, J. Clin. Invest. 118 (2008) 3378–3389.
[39] G. Li, M. Mongillo, K.T. Chin, H. Harding, D. Ron, A.R. Marks, I. Tabas, Role of ERO1-alpha-mediated stimulation of inositol 1,4,5-triphosphate receptor activity in endoplasmic reticulum stress-induced apoptosis, J. Cell Biol. 186 (2009) 783–792.
[40] Y. Tsujimoto, T. Nakagawa, S. Shimizu, Mitochondrial membrane permeability transition and cell death, Biochim. Biophys. Acta 1757 (2006) 1297–1300.
[41] 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.
[42] R.M. Silva, V. Ries, T.F. Oo, O. Yarygina, V. Jackson-Lewis, E.J. Ryu, P.D. Lu, S.J. Marciniak, D. Ron, S. Przedborski, N. Kholodilov, L.A. Greene, R.E. Burke, CHOP/GADD153 is a mediator of apoptotic death in substantia nigra dopamine neurons in an in vivo neurotoxin model of parkinsonism, J. Neurochem. 95 (2005) 974–986.
[43] H. Tsukano, T. Gotoh, M. Endo, K. Miyata, H. Tazume, T. Kadomatsu, M. Yano, T. Iwawaki, K. Kohno, K. Araki, H. Mizuta, Y. Oike, The endoplasmic reticulum stress-C/EBP homologous protein pathway-mediated apoptosis in macrophages contributes to the instability of atherosclerotic plaques, Arterioscler. Thromb. Vasc. Biol. 30 (2010) 1925–1932.
[44] C. Ji, R. Mehrian-Shai, C. Chan, Y.H. Hsu, N. Kaplowitz, Role of CHOP in hepatic apoptosis in the murine model of intragastric ethanol feeding, Alcohol. Clin. Exp. Res. 29 (2005) 1496–1503.
[45] J.H. Lin, H. Li, D. Yasumura, H.R. Cohen, C. Zhang, B. Panning, K.M. Shokat, M.M. Lavail, P. Walter, IRE1 signaling affects cell fate during the unfolded protein response, Science 318 (2007) 944–949.
[46] D. Han, A.G. Lerner, L. Vande Walle, J.P. Upton, W. Xu, A. Hagen, B.J. Backes, S.A. Oakes, F.R. Papa, IRE1alpha kinase activation modes control alternate endoribonuclease outputs to determine divergent cell fates, Cell 138 (2009) 562–575.
[47] H. Nishitoh, A. Matsuzawa, K. Tobiume, K. Saegusa, K. Takeda, K. Inoue, S. Hori, A. Kakizuka, H. Ichijo, ASK1 is essential for endoplasmic reticulum stress-induced neuronal cell death triggered by expanded polyglutamine repeats, Genes Dev. 16 (2002) 1345–1355.
[48] F. Urano, X. Wang, A. Bertolotti, Y. Zhang, P. Chung, H.P. Harding, D. Ron, Coupling of stress in the ER to activation of JNK protein kinases by transmembrane protein kinase IRE1, Science 287 (2000) 664–666.
[49] K. Castillo, D. Rojas-Rivera, F. Lisbona, B. Caballero, M. Nassif, F.A. Court, S. Schuck, C. Ibar, P. Walter, J. Sierralta, A. Glavic, C. Hetz, BAX inhibitor-1 regulates autophagy by controlling the IRE1alpha branch of the unfolded protein response, EMBO J. 30 (2011) 4465–4478.
[50] P. Hu, Z. Han, A.D. Couvillon, R.J. Kaufman, J.H. Exton, Autocrine tumor necrosis factor alpha links endoplasmic reticulum stress to the membrane death receptor pathway through IRE1alpha-mediated NF-kappaB activation and down-regulation of TRAF2 expression, Mol. Cell. Biol. 26 (2006) 3071–3084.
[51] J. Hollien, J.H. Lin, H. Li, N. Stevens, P. Walter, J.S. Weissman, Regulated Ire1-dependent decay of messenger RNAs in mammalian cells, J. Cell Biol. 186 (2009) 323–331.
[52] J. Hollien, J.S. Weissman, Decay of endoplasmic reticulum-localized mRNAs during the unfolded protein response, Science 313 (2006) 104–107.
[53] M. Bouchecareilh, A. Higa, S. Fribourg, M. Moenner, E. Chevet, Peptides derived from the bifunctional kinase/RNase enzyme IRE1alpha modulate IRE1alpha activity and protect cells from endoplasmic reticulum stress, FASEB J. 25 (2011) 3115–3129.
[54] B.C. Cross, P.J. Bond, P.G. Sadowski, B.K. Jha, J. Zak, J.M. Goodman, R.H. Silverman, T.A. Neubert, I.R. Baxendale, D. Ron, H.P. Harding, The molecular basis for selective inhibition of unconventional mRNA splicing by an IRE1-binding small molecule, Proc. Natl. Acad. Sci. U. S. A. 109 (2012) E869–E878.
[55] J.P. Upton, L. Wang, D. Han, E.S. Wang, N.E. Huskey, L. Lim, M. Truitt, M.T. McManus, D. Ruggero, A. Goga, F.R. Papa, S.A. Oakes, IRE1alpha cleaves select microRNAs during ER stress to derepress translation of proapoptotic caspase-2, Science 338 (2012) 818–822.
[56] A.G. Lerner, J.P. Upton, P.V. Praveen, R. Ghosh, Y. Nakagawa, A. Igbaria, S. Shen, V. Nguyen, B.J. Backes, M. Heiman, N. Heintz, P. Greengard, S. Hui, Q. Tang, A. Trusina, S.A. Oakes, F.R. Papa, IRE1alpha induces thioredoxin-interacting protein to activate the NLRP3 inflammasome and promote programmed cell death under irremediable ER stress, Cell Metab. 16 (2012) 250–264.
[57] C. Hetz, L.H. Glimcher, Fine-tuning of the unfolded protein response: assembling the IRE1alpha interactome, Mol. Cell 35 (2009) 551–561.
[58] C. Hetz, F. Martinon, D. Rodriguez, L.H. Glimcher, The unfolded protein response: integrating stress signals through the stress sensor IRE1alpha, Physiol. Rev. 91 (2011) 1219–1243.
[59] R.J. Youle, A. Strasser, The BCL-2 protein family: opposing activities that mediate cell death, Nat. Rev. Mol. Cell Biol. 9 (2008) 47–59.
[60] S. Cory, J.M. Adams, The Bcl2 family: regulators of the cellular life-or-death switch, Nat. Rev. Cancer 2 (2002) 647–656.
[61] G. Chinnadurai, S. Vijayalingam, S.B. Gibson, BNIP3 subfamily BH3-only proteins: mitochondrial stress sensors in normal and pathological functions, Oncogene 27 (Suppl. 1) (2008) S114–S127.
[62] H.M. Zhang, P. Cheung, B. Yanagawa, B.M. McManus, D.C. Yang, BNips: a group of pro-apoptotic proteins in the Bcl-2 family, Apoptosis 8 (2003) 229–236.
[63] S.W. Tait, D.R. Green, Mitochondria and cell death: outer membrane permeabilization and beyond, Nat. Rev. Mol. Cell Biol. 11 (2010) 621–632.
[64] A. Letai, M.C. Bassik, L.D. Walensky, M.D. Sorcinelli, S. Weiler, S.J. Korsmeyer, Distinct BH3 domains either sensitize or activate mitochondrial apoptosis, serving as prototype cancer therapeutics, Cancer Cell 2 (2002) 183–192.
[65] C. Hetz, L. Glimcher, The daily job of night killers: alternative roles of the BCL-2 family in organelle physiology, Trends Cell Biol. 18 (2008) 38–44.
[66] C.A. Hetz, ER stress signaling and the BCL-2 family of proteins: from adaptation to irreversible cellular damage, Antioxid. Redox Signal. 9 (2007) 2345–2355.
[67] E. Szegezdi, D.C. Macdonald, T. Ni Chonghaile, S. Gupta, A. Samali, Bcl-2 family on guard at the ER, Am. J. Physiol. Cell Physiol. 296 (2009) C941–C953.
[68] C.C. Jiang, D. Wroblewski, F. Yang, P. Hersey, X.D. Zhang, Human melanoma cells under endoplasmic reticulum stress are more susceptible to apoptosis induced by the BH3 mimetic obatoclax, Neoplasia 11 (2009) 945–955.
[69] C. Hetz, P. Bernasconi, J. Fisher, A.H. Lee, M.C. Bassik, B. Antonsson, G.S. Brandt, N.N. Iwakoshi, A. Schinzel, L.H. Glimcher, S.J. Korsmeyer, Proapoptotic BAX and BAK modulate the unfolded protein response by a direct interaction with IRE1alpha, Science 312 (2006) 572–576.
[70] D.A. Rodriguez, S. Zamorano, F. Lisbona, D. Rojas-Rivera, H. Urra, J.R. Cubillos-Ruiz, R. Armisen, D.R. Henriquez, E.H. Cheng, M. Letek, T. Vaisar, T. Irrazabal, C. Gonzalez-Billault, A. Letai, F.X. Pimentel-Muinos, G. Kroemer, C. Hetz, BH3-only proteins are part of a regulatory network that control the sustained signalling of the unfolded protein response sensor IRE1alpha, EMBO J. 31 (2012) 2322–2335.
[71] L. Dong, C.C. Jiang, R.F. Thorne, A. Croft, F. Yang, H. Liu, C.E. de Bock, P. Hersey, X.D. Zhang, Ets-1 mediates upregulation of Mcl-1 downstream of XBP-1 in human melanoma cells upon ER stress, Oncogene 30 (2011) 3716–3726.
[72] C.C. Jiang, K. Lucas, K.A. Avery-Kiejda, M. Wade, C.E. deBock, R.F. Thorne, J. Allen, P. Hersey, X.D. Zhang, Up-regulation of Mcl-1 is critical for survival of human melanoma cells upon endoplasmic reticulum stress, Cancer Res. 68 (2008) 6708–6717.
[73] S. Varadarajan, E.T. Bampton, J.L. Smalley, K. Tanaka, R.E. Caves, M. Butterworth, J. Wei, M. Pellecchia, J. Mitcheson, T.W. Gant, D. Dinsdale, G.M. Cohen, A novel cellular stress response characterised by a rapid reorganisation of membranes of the endoplasmic reticulum, Cell Death Differ. 19 (2012) 1896–1907.
[74] A.P. Ghosh, B.J. Klocke, M.E. Ballestas, K.A. Roth, CHOP potentially co-operates with FOXO3a in neuronal cells to regulate PUMA and BIM expression in response to ER stress, PLoS One 7 (2012) e39586.
[75] N. Morishima, K. Nakanishi, K. Tsuchiya, T. Shibata, E. Seiwa, Translocation of Bim to the endoplasmic reticulum (ER) mediates ER stress signaling for activation of caspase-12 during ER stress-induced apoptosis, J. Biol. Chem. 279 (2004) 50375–50381.
[76] J. Li, B. Lee, A.S. Lee, Endoplasmic reticulum stress-induced apoptosis: multiple pathways and activation of p53-up-regulated modulator of apoptosis (PUMA) and NOXA by p53, J. Biol. Chem. 281 (2006) 7260–7270.
[77] J.L. Armstrong, R. Flockhart, G.J. Veal, P.E. Lovat, C.P. Redfern, Regulation of endoplasmic reticulum stress-induced cell death by ATF4 in neuroectodermal tumor cells, J. Biol. Chem. 285 (2010) 6091–6100.
[78] W. Elyaman, F. Terro, K.C. Suen, C. Yardin, R.C. Chang, J. Hugon, BAD and Bcl-2 regulation are early events linking neuronal endoplasmic reticulum stress to mitochondria-mediated apoptosis, Brain Res. Mol. Brain Res. 109 (2002) 233–238.
[79] S. Gupta, L. Cuffe, E. Szegezdi, S.E. Logue, C. Neary, S. Healy, A. Samali, Mechanisms of ER stress-mediated mitochondrial membrane permeabilization, Int. J. Cell Biol. 2010 (2010) 170215.

[80] J.P. Upton, K. Austgen, M. Nishino, K.M. Coakley, A. Hagen, D. Han, F.R. Papa, S.A. Oakes, Caspase-2 cleavage of BID is a critical apoptotic signal downstream of endoplasmic reticulum stress, Mol. Cell. Biol. 28 (2008) 3943–3951.

[81] H. Urra, C. Hetz, The ER in 4D: a novel stress pathway controlling endoplasmic reticulum membrane remodeling, Cell Death Differ. 19 (2012) 1893–1895.

[82] M. Klee, F.X. Pimentel-Muinos, Bcl-X_L specifically activates Bak to induce swelling and restructuring of the endoplasmic reticulum, J. Cell Biol. 168 (2005) 723–734.

[83] N. Echeverry, D. Bachmann, F. Ke, A. Strasser, H.U. Simon, T. Kaufmann, Intracellular localization of the BCL-2 family member BOK and functional implications, Cell Death Differ. 20 (2013) 785–799.

[84] K. Nakajima, H. Hirose, M. Taniguchi, H. Kurashina, K. Arasaki, M. Nagahama, K. Tani, A. Yamamoto, M. Tagaya, Involvement of BNIP1 in apoptosis and endoplasmic reticulum membrane fusion, EMBO J. 23 (2004) 3216–3226.

[85] S.W. Ryu, K. Choi, J. Yoon, S. Kim, C. Choi, Endoplasmic reticulum-specific BH3-only protein BNIP1 induces mitochondrial fragmentation in a Bcl-2- and Drp1-dependent manner, J. Cell. Physiol. 227 (2012) 3027–3035.

[86] A.H. Chaanine, D. Jeong, L. Liang, E.R. Chemaly, K. Fish, R.E. Gordon, R.J. Hajjar, JNK modulates FOXO3a for the expression of the mitochondrial death and mitophagy marker BNIP3 in pathological hypertrophy and in heart failure, Cell Death Dis. 3 (2012) 265.

[87] A.H. Chaanine, R.E. Gordon, E. Kohlbrenner, L. Benard, D. Jeong, R.J. Hajjar, Potential role of BNIP3 in cardiac remodeling, myocardial stiffness and endoplasmic reticulum—mitochondrial calcium homeostasis in diastolic and systolic heart failure, Circ. Heart Fail. 6 (2013) 572–583.

[88] C. Hetz, E. Chevet, H.P. Harding, Targeting the unfolded protein response in disease, Nat. Rev. Drug Discov. 12 (2013) 703–719.

[89] K.Y. Soo, J.D. Atkin, M. Farg, A.K. Walker, M.K. Horne, P. Nagley, Bim links ER stress and apoptosis in cells expressing mutant SOD1 associated with amyotrophic lateral sclerosis, PLoS One 7 (2012) e35413.

[90] H. Suzuki, K. Lee, M. Matsuoka, TDP-43-induced death is associated with altered regulation of BIM and Bcl-xL and attenuated by caspase-mediated TDP-43 cleavage, J. Biol. Chem. 286 (2011) 13171–13183.

[91] R. Leon, N. Bhagavatula, O. Ulukpo, M. McCollum, J. Wei, BimEL as a possible molecular link between proteasome dysfunction and cell death induced by mutant huntingtin, Eur. J. Neurosci. 31 (2010) 1915–1925.

[92] Z. Galehdar, P. Swan, B. Fuerth, S.M. Callaghan, D.S. Park, S.P. Cregan, Neuronal apoptosis induced by endoplasmic reticulum stress is regulated by ATF4–CHOP-mediated induction of the Bcl-2 homology 3-only member PUMA, J. Neurosci. 30 (2010) 16938–16948.

[93] D. Kieran, I. Woods, A. Villunger, A. Strasser, J.H. Prehn, Deletion of the BH3-only protein puma protects motoneurons from ER stress-induced apoptosis and delays motoneuron loss in ALS mice, Proc. Natl. Acad. Sci. U. S. A. 104 (2007) 20606–20611.

[94] P. Nickson, A. Toth, P. Erhardt, PUMA is critical for neonatal cardiomyocyte apoptosis induced by endoplasmic reticulum stress, Cardiovasc. Res. 73 (2007) 48–56.

[95] S. Kiviluoto, T. Vervliet, H. Ivanova, J.P. Decuypere, H. De Smedt, L. Missiaen, G. Bultynck, J.B. Parys, Regulation of inositol 1,4,5-trisphosphate receptors during endoplasmic reticulum stress, Biochim. Biophys. Acta (2013).

[96] D. Prins, M. Michalak, Organellar calcium buffers, Cold Spring Harb. Perspect. Biol. 3 (2011).

[97] H. Coe, M. Michalak, Calcium binding chaperones of the endoplasmic reticulum, Gen. Physiol. Biophys. 28 (2009) F96–F103(Spec No Focus).

[98] D. Rojas-Rivera, R. Armisen, A. Colombo, G. Martinez, A.L. Eguiguren, A. Diaz, S. Kiviluoto, D. Rodriguez, M. Patron, R. Rizzuto, G. Bultynck, M.L. Concha, J. Sierralta, A. Stutzin, C. Hetz, TMBIM3/GRINA is a novel unfolded protein response (UPR) target gene that controls apoptosis through the modulation of ER calcium homeostasis, Cell Death Differ. 19 (2012) 1013–1026.

[99] S.A. Oakes, L. Scorrano, J.T. Opferman, M.C. Bassik, M. Nishino, T. Pozzan, S.J. Korsmeyer, Proapoptotic BAX and BAK regulate the type 1 inositol trisphosphate receptor and calcium leak from the endoplasmic reticulum, Proc. Natl. Acad. Sci. U. S. A. 102 (2005) 105–110.

[100] M. Hammadi, A. Oulidi, F. Gackiere, M. Katsogiannou, C. Slomianny, M. Roudbaraki, E. Dewailly, P. Delcourt, G. Lepage, S. Lotteau, S. Ducreux, N. Prevarskaya, F. Van Coppenolle, Modulation of ER stress and apoptosis by endoplasmic reticulum calcium leak via translocon during unfolded protein response: involvement of GRP78, FASEB J. 27 (2013) 1600–1609.

[101] A. Deniaud, O. Sharaf el dein, E. Maillier, D. Poncet, G. Kroemer, C. Lemaire, C. Brenner, Endoplasmic reticulum stress induces calcium-dependent permeability transition, mitochondrial outer membrane permeabilization and apoptosis, Oncogene 27 (2008) 285–299.

[102] L. Scorrano, S.A. Oakes, J.T. Opferman, E.H. Cheng, M.D. Sorcinelli, T. Pozzan, S.J. Korsmeyer, BAX and BAK regulation of endoplasmic reticulum Ca^{2+}: a control point for apoptosis, Science 300 (2003) 135–139.

[103] R. Bravo, T. Gutierrez, F. Paredes, D. Gatica, A.E. Rodriguez, Z. Pedrozo, M. Chiong, V. Parra, A.F. Quest, B.A. Rothermel, S. Lavandero, Endoplasmic reticulum: ER stress regulates mitochondrial bioenergetics, Int. J. Biochem. Cell Biol. 44 (2012) 16–20.

[104] R. Bravo, J.M. Vicencio, V. Parra, R. Troncoso, J.P. Munoz, M. Bui, C. Quiroga, A.E. Rodriguez, H.E. Verdejo, J. Ferreira, M. Iglewski, M. Chiong, T. Simmen, A. Zorzano, J.A. Hill, B.A. Rothermel, G. Szabadkai, S. Lavandero, Increased ER–mitochondrial coupling promotes mitochondrial respiration and bioenergetics during early phases of ER stress, J. Cell Sci. 124 (2011) 2143–2152.

[105] M.C. Bassik, L. Scorrano, S.A. Oakes, T. Pozzan, S.J. Korsmeyer, Phosphorylation of BCL-2 regulates ER Ca^{2+} homeostasis and apoptosis, EMBO J. 23 (2004) 1207–1216.

[106] S.A. Oakes, J.T. Opferman, T. Pozzan, S.J. Korsmeyer, L. Scorrano, Regulation of endoplasmic reticulum Ca^{2+} dynamics by proapoptotic BCL-2 family members, Biochem. Pharmacol. 66 (2003) 1335–1340.

[107] M. Lam, G. Dubyak, L. Chen, G. Nunez, R.L. Miesfeld, C.W. Distelhorst, Evidence that BCL-2 represses apoptosis by regulating endoplasmic reticulum-associated Ca^{2+} fluxes, Proc. Natl. Acad. Sci. U. S. A. 91 (1994) 6569–6573.

[108] C.W. Distelhorst, M. Lam, T.S. McCormick, Bcl-2 inhibits hydrogen peroxide-induced ER Ca^{2+} pool depletion, Oncogene 12 (1996) 2051–2055.

[109] P. Pinton, D. Ferrari, P. Magalhaes, K. Schulze-Osthoff, F. Di Virgilio, T. Pozzan, R. Rizzuto, Reduced loading of intracellular Ca^{2+} stores and downregulation of capacitative Ca^{2+} influx in Bcl-2-overexpressing cells, J. Cell Biol. 148 (2000) 857–862.

[110] R. Foyouzi-Youssefi, S. Arnaudeau, C. Borner, W.L. Kelley, J. Tschopp, D.P. Lew, N. Demaurex, K.H. Krause, Bcl-2 decreases the free Ca^{2+} concentration within the endoplasmic reticulum, Proc. Natl. Acad. Sci. U. S. A. 97 (2000) 5723–5728.

[111] P. Pinton, D. Ferrari, E. Rapizzi, F. Di Virgilio, T. Pozzan, R. Rizzuto, The Ca^{2+} concentration of the endoplasmic reticulum is a key determinant of ceramide-induced apoptosis: significance for the molecular mechanism of Bcl-2 action, EMBO J. 20 (2001) 2690–2701.

[112] S.A. Oakes, S.S. Lin, M.C. Bassik, The control of endoplasmic reticulum-initiated apoptosis by the BCL-2 family of proteins, Curr. Mol. Med. 6 (2006) 99–109.

[113] 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 1787 (2009) 1342–1351.

[114] W.X. Zong, C. Li, G. Hatzivassiliou, T. Lindsten, Q.C. Yu, J. Yuan, C.B. Thompson, Bax and Bak can localize to the endoplasmic reticulum to initiate apoptosis, J. Cell Biol. 162 (2003) 59–69.

[115] R. Chen, I. Valencia, F. Zhong, K.S. McColl, H.L. Roderick, M.D. Bootman, M.J. Berridge, S.J. Conway, A.B. Holmes, G.A. Mignery, P. Velez, C.W. Distelhorst, Bcl-2 functionally interacts with inositol 1,4,5-trisphosphate receptors to regulate calcium release from the ER in response to inositol 1,4,5-trisphosphate, J. Cell Biol. 166 (2004) 193–203.

[116] 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-X_L modulation of the InsP3R, Nat. Cell Biol. 7 (2005) 1021–1028.

[117] Y.T. Hsu, K.G. Wolter, R.J. Youle, Cytosol-to-membrane redistribution of Bax and Bcl-X_L during apoptosis, Proc. Natl. Acad. Sci. U. S. A. 94 (1997) 3668–3672.

[118] S. Zamorano, D. Rojas-Rivera, F. Lisbona, V. Parra, F.A. Court, R. Villegas, E.H. Cheng, S.J. Korsmeyer, S. Lavandero, C. Hetz, A BAX/BAK and cyclophilin D-independent intrinsic apoptosis pathway, PLoS One 7 (2012) e37782.

[119] A.C. Schinzel, O. Takeuchi, Z. Huang, J.K. Fisher, Z. Zhou, J. Rubens, C. Hetz, N.N. Danial, M.A. Moskowitz, S.J. Korsmeyer, Cyclophilin D is a component of mitochondrial permeability transition and mediates neuronal cell death after focal cerebral ischemia, Proc. Natl. Acad. Sci. U. S. A. 102 (2005) 12005–12010.

[120] S.J. Riedl, Y. Shi, Molecular mechanisms of caspase regulation during apoptosis, Nat. Rev. Mol. Cell Biol. 5 (2004) 897–907.

[121] T. Nakagawa, H. Zhu, N. Morishima, E. Li, J. Xu, B.A. Yankner, J. Yuan, Caspase-12 mediates endoplasmic-reticulum-specific apoptosis and cytotoxicity by amyloid-beta, Nature 403 (2000) 98–103.

[122] J. Hitomi, T. Katayama, Y. Eguchi, T. Kudo, M. Taniguchi, Y. Koyama, T. Manabe, S. Yamagishi, Y. Bando, K. Imaizumi, Y. Tsujimoto, M. Tohyama, Involvement of caspase-4 in endoplasmic reticulum stress-induced apoptosis and Abeta-induced cell death, J. Cell Biol. 165 (2004) 347–356.

[123] F. Yukioka, S. Matsuzaki, K. Kawamoto, Y. Koyama, J. Hitomi, T. Katayama, M. Tohyama, Presenilin-1 mutation activates the signaling pathway of caspase-4 in endoplasmic reticulum stress-induced apoptosis, Neurochem. Int. 52 (2008) 683–687.

[124] T. Yoneda, K. Imaizumi, K. Oono, D. Yui, F. Gomi, T. Katayama, M. Tohyama, Activation of caspase-12, an endoplastic reticulum (ER) resident caspase, through tumor necrosis factor receptor-associated factor 2-dependent mechanism in response to the ER stress, J. Biol. Chem. 276 (2001) 13935–13940.

[125] T. Nakagawa, J. Yuan, Cross-talk between two cysteine protease families. Activation of caspase-12 by calpain in apoptosis, J. Cell Biol. 150 (2000) 887–894.

[126] J.A. Martinez, Z. Zhang, S.I. Svetlov, R.L. Hayes, K.K. Wang, S.F. Larner, Calpain and caspase processing of caspase-12 contribute to the ER stress-induced cell death pathway in differentiated PC12 cells, Apoptosis 15 (2010) 1480–1493.

[127] S. Matsuzaki, T. Hiratsuka, R. Kuwahara, T. Katayama, M. Tohyama, Caspase-4 is partially cleaved by calpain via the impairment of Ca^{2+} homeostasis under the ER stress, Neurochem. Int. 56 (2010) 352–356.

[128] K. Labbe, J. Miu, G. Yeretssian, L. Serghides, M. Tam, C.A. Finney, L.K. Erdman, M.L. Goulet, K.C. Kain, M.M. Stevenson, M. Saleh, Caspase-12 dampens the immune response to malaria independently of the inflammasome by targeting NF-kappaB signaling, J. Immunol. 185 (2010) 5495–5502.

[129] M. Saleh, J.P. Vaillancourt, R.K. Graham, M. Huyck, S.M. Srinivasula, E.S. Alnemri, M.H. Steinberg, V. Nolan, C.T. Baldwin, R.S. Hotchkiss, T.G. Buchman, B.A. Zehnbauer, M.R. Hayden, L.A. Farrer, S. Roy, D.W. Nicholson, Differential modulation of endotoxin responsiveness by human caspase-12 polymorphisms, Nature 429 (2004) 75–79.

[130] M. Kalai, M. Lamkanfi, G. Denecker, M. Boogmans, S. Lippens, A. Meeus, W. Declercq, P. Vandenabeele, Regulation of the expression and processing of caspase-12, J. Cell Biol. 162 (2003) 457–467.

[131] A.M. Scott, M. Saleh, The inflammatory caspases: guardians against infections and sepsis, Cell Death Differ. 14 (2007) 23–31.

[132] A. Jimbo, E. Fujita, Y. Kouroku, J. Ohnishi, N. Inohara, K. Kuida, K. Sakamaki, S. Yonehara, T. Momoi, ER stress induces caspase-8 activation, stimulating cytochrome c release and caspase-9 activation, Exp. Cell Res. 283 (2003) 156–166.

[133] A. Yamamuro, T. Kishino, Y. Ohshima, Y. Yoshioka, T. Kimura, A. Kasai, S. Maeda, Caspase-4 directly activates caspase-9 in endoplasmic reticulum stress-induced apoptosis in SH-SY5Y cells, J. Pharmacol. Sci. 115 (2011) 239–243.

[134] F. Binet, S. Chiasson, D. Girard, Evidence that endoplasmic reticulum (ER) stress and caspase-4 activation occur in human neutrophils, Biochem. Biophys. Res. Commun. 391 (2010) 18–23.
[135] H.H. Cheung, N. Lynn Kelly, P. Liston, R.G. Korneluk, Involvement of caspase-2 and caspase-9 in endoplasmic reticulum stress-induced apoptosis: a role for the IAPs, Exp. Cell Res. 312 (2006) 2347–2357.
[136] H. Gu, X. Chen, G. Gao, H. Dong, Caspase-2 functions upstream of mitochondria in endoplasmic reticulum stress-induced apoptosis by bortezomib in human myeloma cells, Mol. Cancer Ther. 7 (2008) 2298–2307.
[137] S.T. Nawrocki, J.S. Carew, K. Dunner Jr., L.H. Boise, P.J. Chiao, P. Huang, J.L. Abbruzzese, D.J. McConkey, Bortezomib inhibits PKR-like endoplasmic reticulum (ER) kinase and induces apoptosis via ER stress in human pancreatic cancer cells, Cancer Res. 65 (2005) 11510–11519.
[138] G. Carrara, N. Saraiva, C. Gubser, B.F. Johnson, G.L. Smith, Six-transmembrane topology for Golgi anti-apoptotic protein (GAAP) and Bax inhibitor 1 (BI-1) provides model for the transmembrane Bax inhibitor-containing motif (TMBIM) family, J. Biol. Chem. 287 (2012) 15896–15905.
[139] H.J. Chae, H.R. Kim, C. Xu, B. Bailly-Maitre, M. Krajewska, S. Krajewski, S. Banares, J. Cui, M. Digicaylioglu, N. Ke, S. Kitada, E. Monosov, M. Thomas, C.L. Kress, J.R. Babendure, R.Y. Tsien, S.A. Lipton, J.C. Reed, BI-1 regulates an apoptosis pathway linked to endoplasmic reticulum stress, Mol. Cell 15 (2004) 355–366.
[140] R. Huckelhoven, BAX Inhibitor-1, an ancient cell death suppressor in animals and plants with prokaryotic relatives, Apoptosis 9 (2004) 299–307.
[141] K. Reimers, C.Y. Choi, V. Bucan, P.M. Vogt, The Bax Inhibitor-1 (BI-1) family in apoptosis and tumorigenesis, Curr. Mol. Med. 8 (2008) 148–156.
[142] S. Shukla, K. Fujita, Q. Xiao, Z. Liao, S. Garfield, S.M. Srinivasula, A shear stress responsive gene product PP1201 protects against Fas-mediated apoptosis by reducing Fas expression on the cell surface, Apoptosis 16 (2011) 162–173.
[143] H. Zhao, A. Ito, S.H. Kimura, N. Yabuta, N. Sakai, M. Ikawa, M. Okabe, Y. Matsuzawa, S. Yamashita, H. Nojima, RECS1 deficiency in mice induces susceptibility to cystic medial degeneration, Genes Genet. Syst. 81 (2006) 41–50.
[144] M. Fernandez, M.F. Segura, C. Sole, A. Colino, J.X. Comella, V. Cena, Lifeguard/neuronal membrane protein 35 regulates Fas ligand-mediated apoptosis in neurons via microdomain recruitment, J. Neurochem. 103 (2007) 190–203.
[145] B. Bailly-Maitre, C. Fondevila, F. Kaldas, N. Droin, F. Luciano, J.E. Ricci, R. Croxton, M. Krajewska, J.M. Zapata, J.W. Kupiec-Weglinski, D. Farmer, J.C. Reed, Cytoprotective gene bi-1 is required for intrinsic protection from endoplasmic reticulum stress and ischemia–reperfusion injury, Proc. Natl. Acad. Sci. U. S. A. 103 (2006) 2809–2814.
[146] F. Lisbona, D. Rojas-Rivera, P. Thielen, S. Zamorano, D. Todd, F. Martinon, A. Glavic, C. Kress, J.H. Lin, P. Walter, J.C. Reed, L.H. Glimcher, C. Hetz, BAX inhibitor-1 is a negative regulator of the ER stress sensor IRE1alpha, Mol. Cell 33 (2009) 679–691.
[147] S. Buttner, D. Ruli, F.N. Vogtle, L. Galluzzi, B. Moitzi, T. Eisenberg, O. Kepp, L. Habernig, D. Carmona-Gutierrez, P. Rockenfeller, P. Laun, M. Breitenbach, C. Khoury, K.U. Frohlich, G. Rechberger, C. Meisinger, G. Kroemer, F. Madeo, A yeast BH3-only protein mediates the mitochondrial pathway of apoptosis, EMBO J. 30 (2011) 2779–2792.
[148] H.J. Chae, N. Ke, H.R. Kim, S. Chen, A. Godzik, M. Dickman, J.C. Reed, Evolutionarily conserved cytoprotection provided by Bax Inhibitor-1 homologs from animals, plants, and yeast, Gene 323 (2003) 101–113.
[149] Q. Xu, J.C. Reed, Bax inhibitor-1, a mammalian apoptosis suppressor identified by functional screening in yeast, Mol. Cell 1 (1998) 337–346.
[150] C. Xu, W. Xu, A.E. Palmer, J.C. Reed, BI-1 regulates endoplasmic reticulum Ca^{2+} homeostasis downstream of Bcl-2 family proteins, J. Biol. Chem. 283 (2008) 11477–11484.
[151] B.C. Westphalen, J. Wessig, F. Leypoldt, S. Arnold, A. Methner, BI-1 protects cells from oxygen glucose deprivation by reducing the calcium content of the endoplasmic reticulum, Cell Death Differ. 12 (2005) 304–306.
[152] M. Kawai-Yamada, Y. Ohori, H. Uchimiya, Dissection of arabidopsis Bax inhibitor-1 suppressing Bax-, hydrogen peroxide-, and salicylic acid-induced cell death, Plant Cell 16 (2004) 21–32.
[153] M. Krajewska, L. Xu, W. Xu, S. Krajewski, C.L. Kress, J. Cui, L. Yang, F. Irie, Y. Yamaguchi, S.A. Lipton, J.C. Reed, Endoplasmic reticulum protein BI-1 modulates unfolded protein response signaling and protects against stroke and traumatic brain injury, Brain Res. 1370 (2011) 227–237.
[154] B. Bailly-Maitre, B.F. Belgardt, S.D. Jordan, B. Coornaert, M.J. von Freyend, A. Kleinridders, J. Mauer, M. Cuddy, C.L. Kress, D. Willmes, M. Essig, B. Hampel, U. Protzer, J.C. Reed, J.C. Bruning, Hepatic Bax inhibitor-1 inhibits IRE1alpha and protects from obesity-associated insulin resistance and glucose intolerance, J. Biol. Chem. 285 (2010) 6198–6207.
[155] F. de Mattia, C. Gubser, M.M. van Dommelen, H.J. Visch, F. Distelmaier, A. Postigo, T. Luyten, J.B. Parys, H. de Smedt, G.L. Smith, P.H. Willems, F.J. van Kuppeveld, Human Golgi antiapoptotic protein modulates intracellular calcium fluxes, Mol. Biol. Cell 20 (2009) 3638–3645.
[156] X. Wang, K.E. Olberding, C. White, C. Li, Bcl-2 proteins regulate ER membrane permeability to luminal proteins during ER stress-induced apoptosis, Cell Death Differ. 18 (2011) 38–47.
[157] M.C. Wei, T. Lindsten, V.K. Mootha, S. Weiler, A. Gross, M. Ashiya, C.B. Thompson, S.J. Korsmeyer, tBID, a membrane-targeted death ligand, oligomerizes BAK to release cytochrome c, Genes Dev. 14 (2000) 2060–2071.
[158] R. Burikhanov, Y. Zhao, A. Goswami, S. Qiu, S.R. Schwarze, V.M. Rangnekar, The tumor suppressor Par-4 activates an extrinsic pathway for apoptosis, Cell 138 (2009) 377–388.
[159] B.G. Hoffstrom, A. Kaplan, R. Letso, R.S. Schmid, G.J. Turmel, D.C. Lo, B.R. Stockwell, Inhibitors of protein disulfide isomerase suppress apoptosis induced by misfolded proteins, Nat. Chem. Biol. 6 (2010) 900–906.
[160] R.C. Lee, R.L. Feinbaum, V. Ambros, The *C. elegans* heterochronic gene lin-4 encodes small RNAs with antisense complementarity to lin-14, Cell 75 (1993) 843–854.
[161] A.E. Byrd, J.W. Brewer, Micro(RNA)managing endoplasmic reticulum stress, IUBMB Life 65 (2013) 373–381.
[162] M. Maurel, E. Chevet, Endoplasmic reticulum stress signaling: the microRNA connection, Am. J .Physiol. Cell Physiol. 304 (2013) C1117–1126.
[163] S. Behrman, D. Acosta-Alvear, P. Walter, A CHOP-regulated microRNA controls rhodopsin expression, J. Cell Biol. 192 (2011) 919–927.
[164] A.E. Byrd, I.V. Aragon, J.W. Brewer, MicroRNA-30c-2* limits expression of proadaptive factor XBP1 in the unfolded protein response, J. Cell Biol. 196 (2012) 689–698.
[165] Q. Duan, X. Wang, W. Gong, L. Ni, C. Chen, X. He, F. Chen, L. Yang, P. Wang, D.W. Wang, ER stress negatively modulates the expression of the miR-199a/214 cluster to regulates tumor survival and progression in human hepatocellular cancer, PLoS One 7 (2012) e31518.
[166] B.H. Dai, L. Geng, Y. Wang, C.J. Sui, F. Xie, R.X. Shen, W.F. Shen, J.M. Yang, microRNA-199a-5p protects hepatocytes from bile acid-induced sustained endoplasmic reticulum stress, Cell Death Dis. 4 (2013) e604.
[167] M. Maurel, N. Dejeans, S. Taouji, E. Chevet, C.F. Grosset, MicroRNA-1291-mediated silencing of IRE1alpha enhances Glypican-3 expression, RNA 19 (2013) 778–788.
[168] S. Gupta, D.E. Read, A. Deepti, K. Cawley, A. Gupta, D. Oommen, T. Verfaillie, S. Matus, M.A. Smith, J.L. Mott, P. Agostinis, C. Hetz, A. Samali, Perk-dependent repression of miR-106b-25 cluster is required for ER stress-induced apoptosis, Cell Death Dis. 3 (2012) e333.
[169] R. Chhabra, R. Dubey, N. Saini, Gene expression profiling indicate role of ER stress in miR-23a~24–2 cluster induced apoptosis in HEK293T cells, RNA Biol. 8 (2011) 648–664.
[170] N.S. Chitnis, D. Pytel, E. Bobrovnikova-Marjon, D. Pant, H. Zheng, N.L. Maas, B. Frederick, J.A. Kushner, L.A. Chodosh, C. Koumenis, S.Y. Fuchs, J.A. Diehl, miR-211 is a prosurvival microRNA that regulates chop expression in a PERK-dependent manner, Mol. Cell 48 (2012) 353–364.
[171] H.P. Harding, Y. Zhang, A. Bertolotti, H. Zeng, D. Ron, Perk is essential for translational regulation and cell survival during the unfolded protein response, Mol. Cell 5 (2000) 897–904.
[172] M.C. Wei, W.X. Zong, E.H. Cheng, T. Lindsten, V. Panoutsakopoulou, A.J. Ross, K.A. Roth, G.R. MacGregor, C.B. Thompson, S.J. Korsmeyer, Proapoptotic BAX and BAK: a requisite gateway to mitochondrial dysfunction and death, Science 292 (2001) 727–730.