

This Review is part of a thematic series on **Endoplasmic Reticulum Stress and Cardiac Diseases**, which includes the following articles:

What Is the Role of ER Stress in the Heart? Introduction and Series Overview [*Circ Res.* 2010;107:15–18]

The Role of Endoplasmic Reticulum Stress in the Progression of Atherosclerosis [*Circ Res.* 2010;107:839–850]

Endoplasmic Reticulum Stress As a Therapeutic Target in Cardiovascular Disease

Biology of Endoplasmic Reticulum Stress in the Heart

Interrelationship Between Cardiac Hypertrophy, Heart Failure, and Chronic Kidney

Disease—Endoplasmic Reticulum

Masafumi Kitakaze, Guest Editor

Endoplasmic Reticulum Stress As a Therapeutic Target in Cardiovascular Disease

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Abstract: Cardiovascular disease constitutes a major and increasing health burden in developed countries.

Although treatments have progressed, the development of novel treatments for patients with cardiovascular diseases remains a major research goal. The endoplasmic reticulum (ER) is the cellular organelle in which protein folding, calcium homeostasis, and lipid biosynthesis occur. Stimuli such as oxidative stress, ischemic insult, disturbances in calcium homeostasis, and enhanced expression of normal and/or folding-defective proteins lead to the accumulation of unfolded proteins, a condition referred to as ER stress. ER stress triggers the unfolded protein response (UPR) to maintain ER homeostasis. The UPR involves a group of signal transduction pathways that ameliorate the accumulation of unfolded protein by increasing ER-resident chaperones, inhibiting protein translation and accelerating the degradation of unfolded proteins. The UPR is initially an adaptive response but, if unresolved, can lead to apoptotic cell death. Thus, the ER is now recognized as an important organelle in deciding cell life and death. There is compelling evidence that the adaptive and proapoptotic pathways of UPR play fundamental roles in the development and progression of cardiovascular diseases, including heart failure, ischemic heart diseases, and atherosclerosis. Thus, therapeutic interventions that target molecules of the UPR component and reduce ER stress will be promising strategies to treat cardiovascular diseases. In this review, we summarize the recent progress in understanding UPR signaling in cardiovascular disease and its related therapeutic potential. Future studies may clarify the most promising molecules to be investigated as targets for cardiovascular diseases. (*Circ Res.* 2010;107:1071-1082.)

Key Words: heart failure ■ ischemic heart diseases ■ atherosclerosis ■ ER stress ■ unfolded protein response

Although the clinical management of heart failure has advanced substantially,¹ and prevention strategies for atherosclerosis focused on managing the established risk factors have progressed markedly,² cardiovascular disease still constitutes a major and increasing health burden in developed countries. Thus, the development of novel treat-

ments for patients with cardiovascular diseases remains a major research priority.

The endoplasmic reticulum (ER) comprises a complex membranous network found in all eukaryotic cells. It plays a crucial role in the folding of secretory and membrane proteins, calcium homeostasis, and lipid biosynthesis.^{3–5} ER

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Non-standard Abbreviations and Acronyms

AMPK	AMP-activated protein kinase
ASK1	apoptosis signal-regulating kinase 1
ATF	activating transcription factor
Bcl-2	B-cell lymphoma 2
CHOP	CCAAT/enhancer binding protein (C/EBP) homologous protein
CREBH	cAMP response element-binding protein H
eIF2α	eukaryotic initiation factor-2 α
ER	endoplasmic reticulum
ERAD	endoplasmic reticulum-associated degradation
ERO1	endoplasmic reticulum oxidoreductin 1
GRP78	glucose-regulated protein 78 kDa
IκB	inhibitor of κ B
I/R	ischemia/reperfusion
IRE1	inositol-requiring protein 1
JNK	c-Jun N-terminal kinase
KO	knockout
MANF	mesencephalic astrocyte-derived neurotrophic factor
PARM-1	prostatic androgen repressed message-1
PBA	phenylbutyrate
PDI	protein disulfide isomerase
PERK	protein kinase-like endoplasmic reticulum kinase
PKA	protein kinase A
ROS	reactive oxygen species
SERCA	sarcoplasmic reticulum calcium-transporting ATPase
TNF	tumor necrosis factor
TRAF2	tumor necrosis factor receptor-associated factor 2
TRB3	tribbles 3
TLR	Toll-like receptor
UPR	unfolded protein response
XBP1	X box-binding protein-1

stress that disrupts ER function can occur in response to a wide variety of cellular stressors that lead to the accumulation of unfolded and misfolded proteins in the ER. Initially, ER transmembrane sensors detect the accumulation of unfolded proteins and activate transcriptional and translational pathways that deal with unfolded and misfolded proteins, known as the unfolded protein response (UPR). However, the failure to relieve prolonged or severe ER stress causes the cell to undergo apoptotic cell death. Recently, adaptive and proapoptotic pathways of UPR have been implicated in the pathophysiology of human diseases, including cardiovascular diseases, neurodegenerative diseases, diabetes mellitus, obesity, and liver diseases.^{3–5} In this review, we summarize the molecular mechanisms of UPR in cardiovascular diseases and possible therapeutic interventions targeting the components involved in the UPR.

Endoplasmic Reticulum

The ER is the cellular organelle for the synthesis and folding of secreted and membrane-bound proteins and the first site of the secretory pathway.^{3–5} Approximately one-third of newly

synthesized proteins are translocated into the ER, where they fold and assemble with the assistance of ER chaperones and oxidoreductases.⁶ The ER lumen constitutes a specialized environment for proper protein folding and assembly.^{3–5} For instance, the ER contains the millimolar concentrations of free calcium within the cell. Glucose-regulated protein 78 kDa (GRP78), which is the ER-located member of the family of heat shock protein 70 molecular chaperones, promotes the folding of hydrophobic regions in polypeptides to the interior in a calcium-dependent manner.⁷ The oxidizing environment in the ER is crucial for the formation of disulfide bonds mediated by protein disulfide isomerase (PDI) and ER oxidoreductin (ERO)1, which serves as the terminal electron acceptor with oxygen.⁸ Reactive oxygen species (ROS) generated as a product of disulfide bond formation in the ER cause oxidative stress and contribute to apoptotic cell death.⁹ As a consequence of this special environment, the ER is highly sensitive to stresses that deplete its ATP or calcium and alter the intraluminal redox status.

Adaptive and Proapoptotic Pathways of UPR

When unfolded proteins accumulate in the ER, 3 ER transmembrane sensors detect them to initiate 3 distinct UPR branches mediated by the following molecules: protein kinase-like ER kinase (PERK), the inositol requiring kinase (IRE)1, and the transcription factor-activating transcription factor (ATF)6 (Figure 1).^{3–5,10} These UPR sensors have N termini in the lumen of the ER and C termini in the cytosol, thereby connecting the ER and cytosol. All 3 sensors have luminal domains that bind to the ER chaperone GRP78 under normal, unstressed conditions.¹¹ However, on ER stress, GRP78 is released from these sensors, permitting their oligomerization and thereby initiating the UPR to deal with accumulated unfolded proteins. However, if the ER stress is prolonged and/or severe, the ER initiates apoptotic cell death signaling.¹² ER sensor proteins including PERK and IRE1 are responsible for both the adaptive and the proapoptotic pathways of UPR.¹²

IRE1 α is the most fundamental ER stress sensor and is conserved from yeast to humans. On ER stress, the dimerization and autophosphorylation of IRE1 elicit an endoribonuclease activity that specifically cleaves the mRNA encoding the transcription factor X-box binding protein (XBP)1.^{3–5} This unconventional splicing reaction is required for the translation of transcriptionally active XBP1. Active (spliced) XBP1 binds to the ER stress response element in the promoters of a wide variety of UPR-target genes whose products help to fold and degrade the proteins.^{3–5} Our recent study demonstrated that spliced XBP1 can regulate the expression of brain natriuretic peptide (BNP), a non-UPR-target gene, through a novel API/CRE-like element in cardiomyocytes.¹³ Interestingly, p85 α , the regulatory subunit of phosphatidylinositol 3-kinase (PI3K), was found to interact with XBP1 and increase the nuclear translocation of XBP1.¹⁴ In ob/ob mice, the interaction between them is lost, resulting in a severe defect in both the translocation of XBP1 and the resolution of ER stress.¹⁴ These findings suggest that non-UPR and UPR genes are regulated by spliced XBP1.

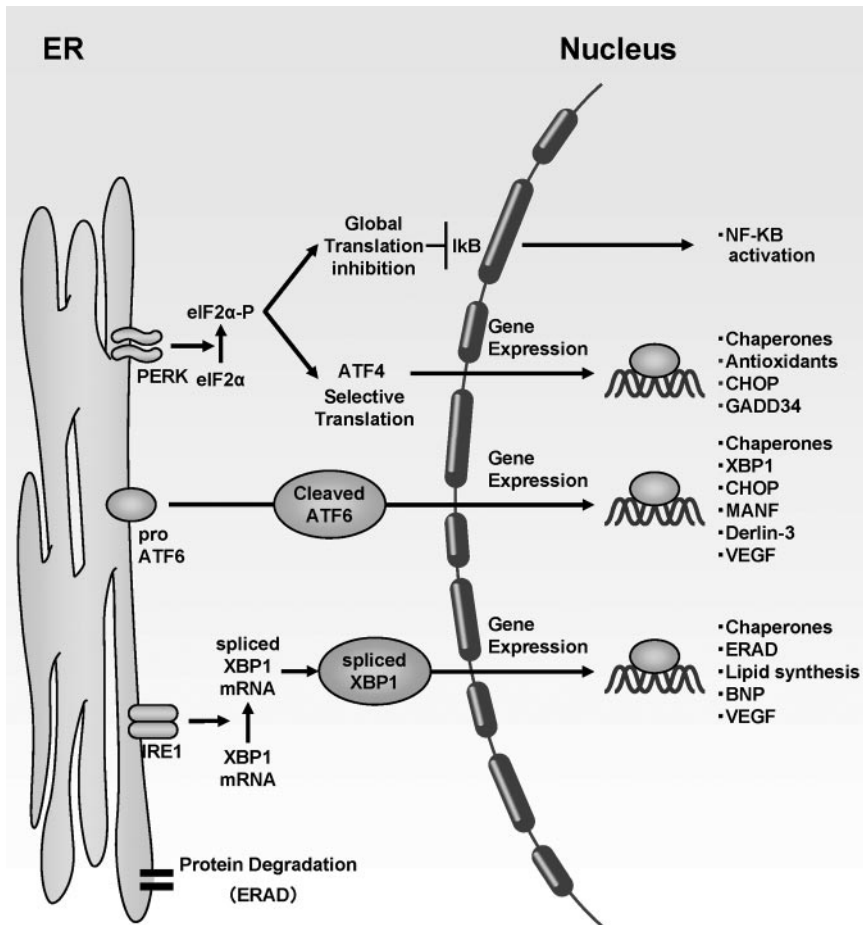


Figure 1. UPR pathways. On ER stress, GRP78 dissociates from 3 ER transmembrane sensors including PERK, IRE1, and ATF6, allowing their activation. PERK phosphorylates eIF2 α , which is dephosphorylated by GADD34, consequently shutting off protein translation. However, paradoxically, eIF2 α phosphorylation induces the selective translation of a transcriptional factor ATF4 to induce the UPR-target genes. ATF6 translocates from ER to Golgi, in which ATF6 cleavage produces a transcriptionally active cytosolic fragment. ATF6 activates a subset of UPR- and non-UPR-target genes, including XBP1. On ER stress, activation of IRE1 elicits an endoribonuclease activity that specifically cleaves the mRNA encoding the transcriptional factor XBP1. This unconventional splicing reaction is required for translation of transcriptionally active XBP1 to induce UPR- and non-UPR-target genes.

In addition to endoribonuclease activity, IRE1 α mediates cell death and inflammatory signaling (Figure 2). IRE1 α interacts with the adaptor protein tumor necrosis factor TNF receptor-associated factor (TRAF) 2, which leads to the activation of a mitogen-activated protein kinase kinase, apoptosis signal-regulating kinase (ASK)1^{15,16} and caspase 12.^{17,18} The ASK1 pathway contributes to the pathogenesis of heart and neurodegenerative diseases.^{19,20} Furthermore, IRE1 α /TRAF2 can also recruit the inhibitor of κ B (I κ B) kinase, which mediates the activation of nuclear factor κ B, suggesting that IRE1 α might provide a link between ER stress and inflammation.²¹

PERK is another ER stress sensor and a serine threonine kinase that phosphorylates eukaryotic translation initiation factor (eIF)2 α on ER stress, resulting in the inhibition of most cap-dependent translation, which requires the interaction of certain key molecules with a special tag bound to the 5'-end of mRNA, termed a cap.³⁻⁵ One example is the translational inhibition of I κ B, resulting in the activation of nuclear factor κ B.²² However, paradoxically, several mRNAs require the phosphorylation of eIF2 α for their translation. One example is the mRNA encoding ATF4, a transcriptional factor that binds to the promoter of the gene encoding GADD34, the regulatory subunit of the phosphatase that dephosphorylates eIF2 α and restores cap-dependent translation.²³ CCAAT/enhancer-binding protein homologous protein (CHOP) is the proapoptotic basic-leucine zipper transcription factor that is

regulated by the ATF4 and ATF6 pathways.³⁻⁵ The deletion of the CHOP gene protects against cell death induced by a pharmacological ER stressor, mechanical stretching and pathophysiological stimuli, such as ischemia and pressure overload.²⁴⁻²⁸ Although the potential mechanisms by which CHOP induces cell death are not well identified, one important pathway by which CHOP induces cell apoptosis is regulation of the balance of proapoptotic and antiapoptotic B-cell lymphoma (Bcl)-2 family proteins. CHOP represses the expression of antiapoptotic proteins Bcl-2 and Bnip3 in cardiomyocytes.^{27,29} In addition, CHOP mediates the direct transcriptional induction and translocation to the ER membrane of Bim, a proapoptotic BH3-only protein of the Bcl-2 protein family, in conditions of ER stress.³⁰

The third ER stress member is ATF6, a transmembrane basic leucine zipper transcription factor.³⁻⁵ ER stress induces the release of GRP78 from ATF6 and permits ATF6 translocation from the ER to the Golgi, where S1P- and S2P-mediated proteolytic cleavage produces a transcriptionally active cytosolic fragment. Cleaved ATF6 binds to the ER stress response element in the promoters of UPR target genes, including XBP1. Recently, several ATF6-related proteins with distinct tissue distributions were identified.^{31,32} Interestingly, lipopolysaccharide and cytokines activate cAMP response element-binding protein (CREB)H, which is a hepatocyte-specific ER-anchored transcription factor that activates a subset of genes associated with inflammation but

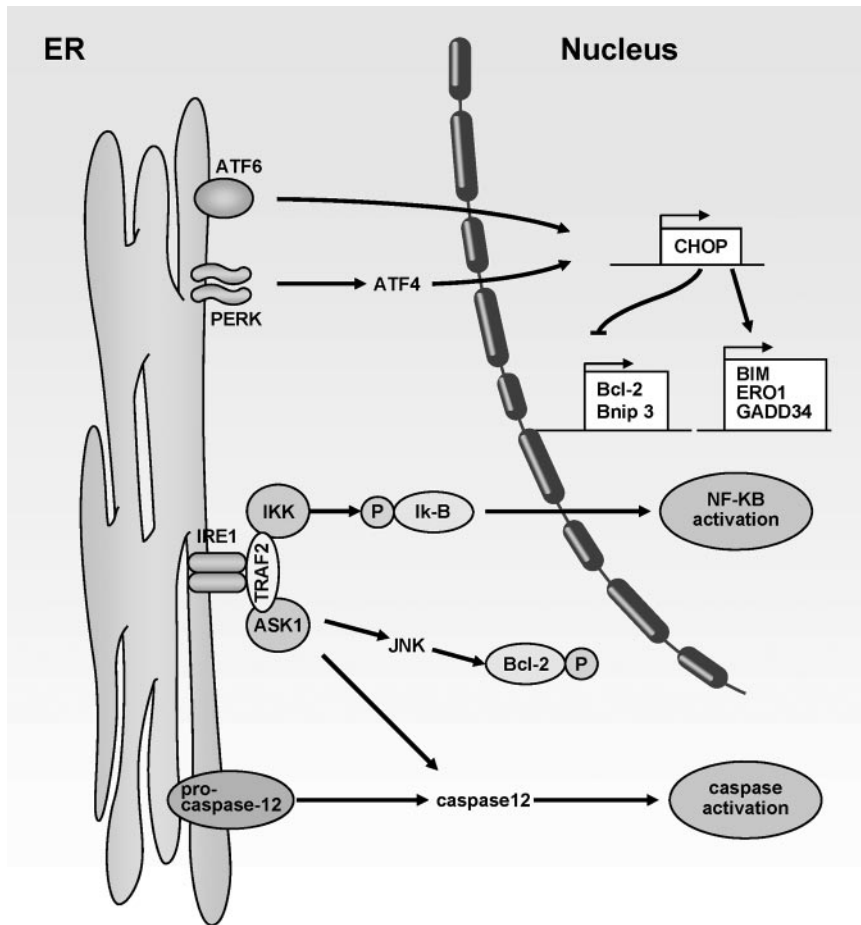


Figure 2. Proapoptotic pathways of UPR. CHOP is the proapoptotic bZIP transcription factor that is regulated mainly by ATF4- and ATF6-dependent pathways. CHOP represses the expression of the antiapoptotic proteins Bcl-2 and Bnip3. In addition, CHOP mediates the direct transcriptional induction and translocation to the ER membrane of Bim, a proapoptotic BH3-only protein of the Bcl-2 family, on ER stress. IRE1 α interacts with the adaptor protein TRAF2. The IRE1 α /TRAF2 complex interacts with ASK1, which subsequently phosphorylates JNK. Activation of JNK induces apoptotic cell death through the phosphorylation of several Bcl-2 family members. The IRE1 α /TRAF2 complex also interacts with I κ B kinase, which leads to nuclear factor κ B (NF- κ B) activation. On ER stress, procaspase 12 is cleaved and activated, which in turn activates caspase-9/3, thereby leading to mitochondria-independent cell death.

not UPR.³¹ These findings suggest that CREBH integrates the UPR with the acute phase response. PARM-1 (prostatic androgen repressed message 1) was shown to be induced in a cardiac hypertrophy and subsequent heart failure model.³³ PARM-1 expression is induced by ER stress, which plays a protective role in cardiomyocytes through the regulation of PERK, ATF6 and CHOP expression. The existence of tissue-specific UPR components allows for the response to tissue-specific stress.

Misfolded ER proteins are exported from the ER into the cytosol by a process termed ER-associated protein degradation (ERAD) or retrotranslocation.^{3,34} Most ERAD substrates are ubiquitinated and extracted by a cytosolic ATPase named p97 before degradation by the proteasome. Defects in ERAD cause the accumulation of misfolded proteins in the ER and thus trigger the UPR.³⁵

ER Stress and Cardiovascular Diseases

Experimentally, the ER environment can be perturbed by substances such as dithiothreitol, thapsigargin, or tunicamycin, which alter the redox status, calcium levels and protein glycosylation in the ER, respectively.³⁶ When cells are treated with one of these compounds, ER protein folding is impaired, and the accumulation of unfolded proteins activates the adaptive and proapoptotic pathways of the UPR.³⁻⁵

Pathophysiological stimuli also activate UPR. Hypoxia, angiotensin II and tumor necrosis factor (TNF)- α activate

adaptive and proapoptotic pathways of the UPR in cultured cardiomyocytes.³⁷⁻³⁹ The cardiac-specific deposition of aggregated β -amyloid⁴⁰ or polyglutamine preamyloid oligomer⁴¹ activated the component of UPR in mouse transgenic hearts. Cyclic stretching significantly increases CHOP protein in cultured cardiomyocytes,²⁸ and CHOP expression increases in pressure-overloaded hearts.³⁸ Metabolic factors such as cholesterol, homocysteine, glucose, fatty acid, and palmitate can also trigger ER stress.^{42,43} These findings suggest that activation of UPR by pathophysiological stimuli is involved in the development of cardiovascular diseases.

Cardiac Hypertrophy and Failure

Electron microscopic analyses have revealed that the proliferation of tubules of the ER is a common finding in degenerated cardiomyocytes, suggesting that the ER is overloaded in this condition.⁴⁴ Oxidative stress, hypoxia, and enhanced protein synthesis found in failing hearts potentially enhance ER stress. Indeed, in patients with heart failure, we and others have shown the existence of spliced XBPI and markedly increased GRP78 expression, suggesting that UPR activation is associated with the pathophysiology of heart failure in humans.^{13,38,45} Our study also showed that mRNA levels of ATF4 and CHOP are increased in these patients.²⁷ Furthermore, ubiquitinated proteins are accumulated in human failing hearts.^{46,47} These findings suggest that protein quality control is impaired in human failing hearts.

In Dahl salt-sensitive rats, a high-salt diet leads to hypertension, cardiac hypertrophy, and subsequent heart failure, as well as significant increases in both GRP78 and CHOP expression.³³ Consistent with these findings, in mice that received transaortic constriction, GRP78 and CHOP are upregulated.³⁸ Interestingly, UPR activation has been found in both hypertrophic and failing hearts, whereas the activation of ER-initiated apoptosis CHOP, but not c-Jun N-terminal kinase (JNK) or caspase 12, is found only in failing hearts.³⁸ These findings suggest that UPR activation is consistently found in hearts subjected to pressure overload. However, when the ER stress is prolonged, the ER-initiated apoptosis signal CHOP is activated in failing mouse hearts induced by pressure overload. To clarify the role of CHOP in hypertrophic and subsequently failing hearts by pressure overload, we performed transaortic constriction using CHOP knockout (KO) mice.²⁷ We observed that both cardiac hypertrophy and dysfunction were attenuated in CHOP KO mice compared with wild type. In CHOP KO mice, the enhanced phosphorylation of eIF2 α , which was attributable to the lack of negative regulation by GADD34, may lead to the global repression of translation. This may explain the mechanism by which cardiac hypertrophy is reduced in pressure-overloaded hearts of CHOP KO mice. Furthermore, microarray analysis revealed that CHOP positively and negatively regulates several proapoptotic and antiapoptotic molecules of the Bcl-2 family. These findings suggest that CHOP might be a promising molecular target for the treatment of cardiac hypertrophy and failure.

Protein accumulation resulting from the impairment of the secretory pathway or mutant protein synthesis also causes heart failure. In transgenic mice systemically expressing a mutant KDEL (Lys-Asp-Glu-Leu) receptor,⁴⁸ which is a retrieval receptor for ER chaperones in the early secretory pathway, disturbed recycling of misfolded proteins between the ER and Golgi complex and enhanced expression of CHOP and apoptosis were observed in the mutant hearts. The transgenic mice exhibited dilated cardiomyopathy without obvious findings in other tissues, suggesting that the heart is sensitive to ER stress. The transgenic expression of mutant proteins in neural and cardiac tissues is a good model to test whether intracellular aggregation affects cardiac function.^{40,41,49,50}

Viruses exploit the translational machinery of the host cell to synthesize their viral proteins, leading to increased folding pressure on the ER and chaperone upregulation.^{51,52} Thus, it is likely that the adaptive and proapoptotic pathways of UPR are involved in the pathophysiology of viral myocarditis. Furthermore, Mao et al demonstrated that ER stress plays an important role in cardiomyocyte apoptosis and the development of dilated cardiomyopathy in rabbits immunized with a peptide corresponding to the β 1-adrenoceptor.^{53,54} These findings suggest that the UPR plays an important role in the pathophysiology of virus and autoimmune heart diseases.

Ischemic Heart Diseases

In a myocardium that has experienced ischemia/reperfusion (I/R), myocardial death and severe inflammation are induced because of the depletion of oxygen and nutrients, followed by

the sudden burden of oxygen free radicals and production of proinflammatory cytokines.⁵⁵ Either of these stimuli can potentially induce the adaptive and proapoptotic pathways of UPR. Indeed, increased expression of UPR-related genes is reported in cardiomyocytes near myocardial infarction in mice and humans.^{56–58}

Martindale et al demonstrated the role of ATF6, a component of the UPR, in I/R injury using transgenic mice with cardiac-restricted expression of a novel tamoxifen-activated form of ATF6.⁵⁹ The tamoxifen-treated transgenic mouse hearts exhibit better functional recovery from ex vivo I/R, as well as significantly reduced necrosis and apoptosis and increased expression of ER-resident chaperones GRP78 and -94. Toko et al demonstrated that the treatment of mice with 4-(2-aminoethyl) benzenesulfonyl fluoride, an inhibitor of ATF6, further reduces cardiac function and increases the mortality rate after myocardial infarction.⁵⁸ These findings suggest that ATF6 exerts cardioprotective effects on I/R injury. Furthermore, Glembotski and colleagues showed that ATF6 activation induces numerous genes in cultured cardiomyocytes, including MANF (mesencephalic astrocyte-derived neurotrophic factor).⁶⁰ Knockdown of endogenous MANF with microRNA increases cell death on simulated I/R, whereas the addition of recombinant MANF to media protected the cultured cardiac myocytes from simulated I/R-mediated death.⁶⁰ It was also shown that activated ATF6 induces the Derlin-3 gene, which encodes an important component of the ERAD machinery. Overexpression of Derlin-3 enhances ERAD and protects cardiomyocytes from simulated ischemia-induced cell death.⁶¹

In rat neonatal cultured cardiomyocytes, hypoxia increased XBP1 mRNA splicing and GRP78 protein levels.⁵⁶ Because infection with a recombinant adenovirus encoding dominant-negative XBP1 augments hypoxia/reoxygenation-induced apoptosis, the XBP1 arm of the UPR may play cardioprotective roles against hypoxic insult. Vitadello et al demonstrated that the overexpression of GRP94, the expression of which is regulated by XBP1 and ATF6, reduces myocyte necrosis caused by calcium overload or simulated ischemia in cardiac H9C2 muscle cells.⁶² In addition, it was reported that ischemic preconditioning or postconditioning reduces cardiac damage associated with UPR activation.^{63,64} In human heart samples, Severino et al demonstrated that PDI is upregulated 3-fold in the viable periinfarct myocardial region.⁶⁵ Adenoviral-mediated PDI gene transfer to the mouse heart results in a 2.5-fold smaller infarct size and significantly reduces cardiomyocyte apoptosis in the periinfarct region and the smaller left ventricular end-diastolic diameter compared with treatment with a transgene-null adenoviral vector.

On the other hand, Terai et al demonstrated that hypoxia induces CHOP expression and the cleavage of caspase 12; this effect is significantly inhibited by pretreatment with a pharmacological activator of AMP-activated protein kinase (AMPK).³⁷ This finding indicates that the proapoptotic pathways of the UPR are involved in cell death by hypoxic stimuli. In addition, Nickson et al demonstrated that ER stress induces the expression of PUMA, a proapoptotic member of the BCL-2 family, and that the suppression of PUMA expression leads to inhibition of cardiomyocyte apoptosis

induced by a pharmacological ER stressor.⁶⁵ Importantly, the targeted deletion of PUMA attenuates cardiomyocyte death and improves cardiac function during I/R periods.⁶⁶ Pharmacological ER stressors can induce tribbles (TRB)3 expression in cultured cardiomyocytes, and myocardial infarction results in cardiac ER stress caused by the induction of TRB3.⁶⁷ Cardiac-specific overexpression of TRB3 sensitizes mice to infarct expansion and cardiomyocyte apoptosis in the infarct border zone after myocardial infarction. Agents that inhibit TRB3 expression or activity may lead to reduced pathological cardiac remodeling in patients.⁶⁷

Furthermore, microbes activate a transcriptional program through Toll-like receptors (TLRs). TLR signaling modifies both the adaptive and the proapoptotic pathways of the UPR.^{68,69} TLR-mediated signaling pathways have been implicated in myocardial I/R injury.^{70,71} Disruption of TLR2-mediated signaling may be helpful for the induction of immediate or delayed myocardial protection from I/R injury.⁷¹ Further investigation should identify the possible involvement of TLR-ER signaling in I/R injury.

Potential Cardiotoxicity of the New Cancer Therapy

In recent years, small-molecule inhibitors of protein kinases, including receptor tyrosine kinase inhibitors, have been developed for cancer treatment. Interestingly, Kerkela et al demonstrated that imatinib, the first tyrosine kinase inhibitor to inhibit the activity of the causal fusion protein Bcr-Abl in chronic myeloid leukemia, induces left ventricular dysfunction in the animal model and in some patients.⁷² In cardiomyocytes, Kerkela et al showed a relationship between c-Abl activity and ER homeostasis, although the mechanism by which c-Abl inhibition destabilizes the ER has not been identified.⁷² This study was the first to show that ER-initiated apoptosis signaling may be involved in the cardiotoxicity of this anticancer drug.⁷³

A proteasome inhibitor is another important advance in molecular anticancer therapy for the treatment of multiple myeloma.⁷³ Because proteasome inhibition causes the accumulation of unfolded proteins, it will activate the UPR. Importantly, treatment with a proteasome inhibitor is associated with a high prevalence of heart failure.⁷⁴ Consistent with that clinical report, we showed that proteasome inhibition induces cardiomyocyte death and activates ER stress-induced transcriptional factor ATF6, but not XBP1, leading to a failure to upregulate ER chaperones.⁷⁵ Overexpression of GRP78 suppresses both CHOP expression and cardiomyocyte death by proteasome inhibition. These findings indicate that proteasome inhibition perturbs ER homeostasis in cardiomyocytes without the induction of ER chaperones, thereby inducing a cycle of cardiac damage. Chemical ER chaperones or drugs that enhance endogenous ER chaperone function in the heart will be promising candidates to prevent cardiotoxicity by proteasome inhibitors,⁷⁶ although it is important to recognize the possibility that chemical chaperones will promote cancer growth.

The UPR was shown to be highly induced in various tumors and closely associated with cancer cell survival.⁷⁷ There are a number of drugs under development that aim to

treat cancer by inhibiting one or more of the branches of the UPR and enhancing ER stress.⁷⁸ Careful monitoring of cardiac function is necessary when using pharmacological agents targeting the UPR.

Atherosclerosis

Advanced atherosclerotic plaques provide a pathophysiological environment that can cause ER stress and activate the UPR; in this environment, oxidized lipids, inflammation, and metabolic stress are induced.⁷⁹ Indeed, ER stress is markedly increased in endothelial cells subjected to atherosclerosis-prone shear stress.⁸⁰ Consistently, transcript profiles revealed that the most abundant feature of the endothelium of all atherosusceptible regions is the upregulation of genes associated with ER stress.⁸¹ Dong et al demonstrated that the dysfunction of AMPK significantly increases the level of ER stress and suppresses sarcoplasmic reticulum calcium-transporting ATPase (SERCA) activity in endothelial cells.⁸² These results suggest that AMPK functions as a physiological suppressor of ER stress by maintaining SERCA activity and intracellular Ca²⁺ homeostasis.

A growing body of evidence indicates that a key cellular event in the conversion of benign to vulnerable atherosclerotic plaques is ER stress-induced macrophage apoptosis.⁸³ Interestingly, macrophage and smooth muscle cells in atherosclerotic plaques produce abundant secretory proteins, which potentially induce ER stress in these cells.⁷⁹ A cause of macrophage death is the accumulation of free cholesterol in the ER, leading to activation of the UPR and CHOP-induced apoptosis.^{84–86} Macrophage fatty acid-binding protein-4 (aP2), a cytosolic lipid chaperone, plays an important role in cellular lipid metabolism and the reception of lipid signals.⁸⁷ Erbay et al demonstrated that aP2 is the predominant regulator of lipid-induced macrophage ER stress and that its absence increases bioactive lipids that render macrophages resistant to lipid-induced ER stress.⁸⁸ Another study shows that impaired proteasome function promotes features of a more rupture-prone plaque phenotype in apolipoprotein (Apo)E KO mice.⁸⁹ These findings suggest that ER stress is closely involved in the development of atherosclerosis and that ER-related molecules are promising candidates for therapy.

We investigated the association between the ER stress and plaque rupture in 152 human coronary artery autopsy samples. We found a strong association between ER stress markers such as CHOP and GRP78 and ruptured atherosclerotic plaques in human coronary artery lesions,⁹⁰ suggesting that ER stress is likely involved in the development of plaque rupture in humans. Importantly, Thorp et al provide direct evidence for a causal link between the ER-initiated apoptosis signaling of CHOP and plaque necrosis.⁹¹ It was shown that plaque necrosis and lesion apoptosis are markedly reduced in the CHOP-deficient mice mated with ApoE KO or low-density lipoprotein receptor KO atherosclerotic mice.⁹¹

Targeting UPR As Potential Therapy in Cardiovascular Diseases

There are 2 main approaches to targeting the UPR for the treatment of cardiovascular diseases. The first approach

Table. Drugs Affecting UPR and ER-Initiated Apoptosis Signaling

Drug	Mechanism of Action	Potential Indication
Phenylbutyrate	Chemical chaperone	Cardioprotection ^{28,119} ; atherosclerosis prevention ⁸⁸
Tauroursodeoxycholic acid	Chemical chaperone	Cardioprotection; atherosclerosis prevention ⁸⁸
BiP inducer X ⁷⁶	GRP78 induction	Cardioprotection
Curcumin ¹⁰⁴	GRP94 induction	Cardioprotection
Salubrinal ⁹³	Prevention of eIF2 α dephosphorylation	Cardioprotection; prevention of cardiac hypertrophy
Sunitinib ⁹⁶	IRE1 activation	
EN460	ERO1 inhibitor	ROS reduction ¹²⁵
Fused heterocyclic compound	ASK1 inhibitor	Cardioprotection ^{4,126}
SP600125	Prevention of CHOP induction by stretch	Prevention of cardiac hypertrophy/failure ²⁸ and atherosclerosis
SB203580	CHOP phosphorylation	Prevention of cardiac hypertrophy/failure ²⁸ and atherosclerosis
TNF- α antibody	Prevention of CHOP induction	Prevention of cardiac hypertrophy/failure ^{28,39} and atherosclerosis
CS-866	Reduction of ER stress by pressure-overload	Cardioprotection ³⁸
Pravastatin	Reduction of ER stress by pressure-overload	Cardioprotection ³⁹
Pioglitazone ¹²⁰	Reduction of ER stress	Cardioprotection; atherosclerosis prevention
AICAR; metformin	Reduction of ER stress by AMPK activation	Cardioprotection ^{37,121} ; atherosclerosis prevention ^{92,122}
Isoproterenol	Proteasome activation and assembly	Cardioprotection ¹⁰⁷ ; atherosclerosis prevention

AICAR indicates 5-aminoimidazole-4-carboxamide-1- β -D-ribofuranoside.

involves activating components of the adaptive pathway of the UPR to deal with the stress. The second approach is to deactivate the components of the proapoptotic pathways of UPR. Although targeting the UPR components seems promising as a potential therapy for cardiovascular diseases, there are several limitations in our knowledge. For example, the ER stress sensors IRE1 α and PERK elicit signaling to maintain ER homeostasis and to induce cell death. Unfortunately, at the present time, the mechanisms by which the signaling switches from cell survival to death are not fully understood. Thus, we cannot know the precise moment to activate or inhibit ER stress sensor proteins for treatment. Although these limitations suggest that it is still at an early stage for potential therapeutic use, the alteration of UPR activation or the reduction of ER stress are still promising therapeutic goals.

Pharmacological Agents Targeting UPR Component

Pharmacological agents that directly activate or deactivate UPR components will be potentially useful in treating cardiovascular diseases (Table). Potential components of the UPR and ERAD, such as ATF6, IRE1, spliced XBP1, PERK, eIF2 α , and the proteasome, could be good initial targets for therapeutic design. For example, activation of ATF6 may exert cardioprotective effects against I/R injury.^{36,56,58–61,92} We have demonstrated that enhanced phosphorylation of eIF2 α may lead to repressed global protein synthesis and attenuated cardiac hypertrophy in pressure-overloaded hearts of CHOP KO mice,²⁷ suggesting that enhanced phosphorylation of eIF2 α would be a novel therapeutic target for the prevention of cardiac hypertrophy. Salubrinal is a small molecule that prevents dephosphorylation of eIF2 α .⁹³ This compound significantly increases the Grp78/Bip ER chaperone levels and results in attenuated caspase-4-dependent apoptosis in A β -treated neurons.^{40,94} Although this chemical

strategy could be beneficial for treating cardiac hypertrophy, we note that different cell types respond differently to salubrinal.⁹⁵ Korennykh et al demonstrated that several kinase inhibitors, including sunitinib, can directly activate IRE1, leading to XBP-1 splicing and a reduction in ER stress.⁹⁶ However, we note that patients receiving sunitinib, especially those with a previous history of hypertension and coronary heart disease, are at increased risk for cardiovascular events and should be monitored for exacerbations of their hypertension and for evidence of LVEF dysfunction during treatment.^{97,98} Thus, it may be of therapeutic value to separate the intended function of kinase inhibitors from the unintended activation of IRE1. Finally, the UPR components are broadly expressed, and augmentation or inhibition of their activity may lead to unintended toxicity if the agent is not selectively delivered to the cell type or organ of interest.⁹⁹ Targeting cell-specific ER component such as CREBH or PARM-1 would be a good strategy.

Although there are no pharmacological agents targeting CHOP, experimental results suggest that the development of CHOP inhibitors would attenuate cardiac hypertrophy and failure and prevent atherosclerosis.^{27,91} CHOP activates ERO1 α , which catalyzes reoxidation of PDI, resulting in the production of hydrogen peroxide.^{100,116} ROS generated as a byproduct of disulfide bond formation in the ER cause oxidative stress and contribute to apoptotic cell death.^{9,101} ERO1 α may thus be an important mediator of apoptosis downstream of CHOP. Ron and colleagues report that the inhibitor EN460 interacts selectively with the reduced, active form of ERO1 α and prevents its reoxidation.¹⁰² This selectivity is explained by the rapid reversibility of the reaction of EN460 with unstructured thiols, in contrast to the formation of a stable bond with ERO1 α . Moreover, both the antioxidant *N*-acetylcysteine and the ER stress inhibitor tauroursodeoxycholic acid reverse the reduced cardiomyocyte contractile function elicited by the oxidative stress inducer menadi-

one.¹⁰³ Curcumin and masoprocol can serve as lead candidate prophylactics for ROS induced ER stress as they preserve the functional integrity of the PDI.¹⁰⁴ Genetic or chemical intervention to reduce ROS levels improves protein folding and cell survival and may provide an avenue to treat and/or prevent cardiovascular diseases.⁹

Pharmacological agents that indirectly target the UPR and ERAD pathways will also be promising. Cyclic stretching significantly increased CHOP protein levels; this increase is reduced by the addition of JNK inhibitor SP600125 or TNF- α antibody 30 minutes before the stretch stimuli.²⁸ An inhibitor of p38 mitogen-activated protein kinase, SB203580, abolished the stress-inducible *in vivo* phosphorylation of CHOP.¹⁰⁵ Thus, these findings suggest that mitogen-activated protein kinase inhibitors may modify components of the UPR pathways. Finally, a fused heterocyclic compound is now under development to inhibit ASK1 activity, a downstream signal of IRE1, because ASK1 is involved in cardiac hypertrophy and failure.^{20,106} Interestingly, we have demonstrated that ischemic preconditioning and activation of protein kinase A with isoproterenol or forskolin enhance proteasome assembly and activity.¹⁰⁷ Because proteasome inhibition promotes atherosclerosis and cardiac dysfunction,^{46,89} protein kinase A activators may attenuate ER-initiated apoptosis signaling through enhanced proteasome function.

Chemical ER Chaperones

GRP78 is the ER homolog of HSP70 proteins with a conserved ATPase domain and a peptide-binding domain.^{7,108} As a chaperone, GRP78 recognizes and binds to proteins with hydrophobic residues in the unfolded regions.^{7,108} GRP94 shares common transcriptional regulatory elements with the GRP78 promoter and is coordinately regulated with GRP78. It is reasonable to speculate that overexpression of ER-resident chaperone aids in the folding of proteins and attenuation of ER stress.¹⁰⁹ Indeed, overexpression of GRP78/94 attenuates ER stress and cardiac damage by I/R⁶² or proteasome inhibition.^{62,75,110} Furthermore, ischemic preconditioning protects hearts against I/R injury associated with the increased expression of GRP78.⁶³ Consistent with this observation, a small chemical induces GRP78 and protects against ER stress in neurons.⁷⁶

One promising approach is the use of pharmacological agents, such as small chemical chaperones, which can stabilize proteins in their native conformation and rescue mutant protein folding and/or trafficking defects.^{111,112} Sodium phenylbutyrate (PBA) is approved for the chronic adjunctive treatment of certain urea cycle disorders.¹¹³ It also functions as a chemical chaperone because of the preferential hydration of the exposed polypeptide backbones and side chains of partially unfolded structures.^{114,115} PBA is now used in the clinical setting for the treatment of diseases associated with protein misfolding, such as α 1-antitrypsin deficiency and cystic fibrosis.^{114,115} Ozcan et al have shown that chemical chaperones reduce ER stress and restore glucose homeostasis in a mouse model of type 2 diabetes.¹¹⁶ Erbay et al have shown that ER modification by chemical chaperones in macrophages and adipocytes has therapeutic efficacy against atherosclerosis in mouse models.⁸⁸ Furthermore, PBA rescues

ER stress-induced suppression of APP proteolysis and prevents apoptosis in neuronal cells¹¹⁷ and inhibits adipogenesis by modulating the UPR.¹¹⁸ Although the authors did not check the UPR pathways, PBA also protects against doxorubicin-induced cardiac injury.¹¹⁹ Because PBA is clinically approved for some diseases,¹¹³ it will be fascinating to test its efficacy in humans with cardiovascular disease. However, we must emphasize that there are several limitations with these chemicals because of the high doses needed to produce the desired effect. Further studies will be required to elucidate how these chemical chaperones promote protein folding and to develop their analogs with desirable pharmacokinetics for the treatment of cardiovascular diseases.

Clinically Approved Pharmaceutical Agents

Some pharmacological agents used in clinical settings may affect UPR pathways. In pressure-overloaded hearts, we found that increased expression of CHOP was partially inhibited by an angiotensin II type 1 receptor antagonist.³⁸ Additionally, cardiac expression of TNF- α is increased in pressure-overloaded hearts, and TNF- α induces ER stress in cultured neonatal rat cardiomyocytes; both of these effects are significantly inhibited by pravastatin.³⁹ The antidiabetic agent pioglitazone suppresses ER stress in the liver, which may in part explain the pharmacological effects of pioglitazone in reducing insulin resistance.¹²⁰

Pharmaceutical agents that activate AMPK are other promising agents. Activation of AMPK reduces cardiac ER stress and prevents the progression of heart failure in dogs.^{37,121} Furthermore, the reduction of AMPK activity increases ER stress and atherosclerosis *in vivo*.^{82,122} Tempol, which restores SERCA activity and decreases oxidized SERCA levels, markedly reduces the level of ER stress in mouse aortic endothelial cells from AMPK α 2-deficient mice. Finally, oral administration of tauroursodeoxycholic acid, a chemical chaperone that inhibits ER stress, significantly reduces both ER stress and aortic lesion development in low-density lipoprotein receptor- and AMPK α 2-deficient mice.^{82,122} These findings suggest that AMPK activators may reduce ER stress associated with the improvement of cardiovascular disease. Activation of the receptor for glucagon-like peptide-1 reduces ER stress in pancreatic β cells.¹²³ Because the glucagon-like peptide-1 receptor is expressed throughout the mouse cardiovascular system,¹²⁴ it will be interesting to investigate its effects on cardiovascular disease.

Concluding Remarks

Although understanding the pathophysiological role of ER stress in cardiovascular diseases has progressed in recent years, future research needs to address several critical questions. (1) To what extent are the adaptive and proapoptotic pathways of the UPR involved in the pathophysiology of cardiovascular disease? (2) If we activate or deactivate components of UPR, will it improve disease condition? (3) Can we really regulate the cell survival and death signal involved in the UPR? (4) How can we deliver the agent to the targeted tissues? The improved understanding of the underlying molecular mechanisms of the UPR in cardiovascular

diseases will provide new targets for drug discovery and therapeutic intervention.

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Disclosures

None.

References

- Tang WHW, Francis GS. The year in heart failure. *J Am Coll Cardiol*. 2010;55:688–696.
- Libby P, Ridker PM, Hansson GK. Inflammation in atherosclerosis: from pathophysiology to practice. *J Am Coll Cardiol*. 2009;54:2129–2138.
- Ron D, Walter P. Signal integration in the endoplasmic reticulum unfolded protein response. *Nat Rev Mol Cell Biol*. 2007;8:519–529.
- Kim I, Xu W, Reed JC. Cell death and endoplasmic reticulum stress: disease relevance and therapeutic opportunities. *Nat Rev Drug Discov*. 2008;7:1013–1030.
- Minamino T, Kitakaze M. ER stress in cardiovascular disease. *J Mol Cell Cardiol*. 2010;48:1105–1110.
- Ghaemmaghami S, Huh WK, Bower K, Howson RW, Belle A, Dephore N, O'Shea EK, Weissman JS. Global analysis of protein expression in yeast. *Nature*. 2003;425:737–741.
- Gething M-J. Role and regulation of the ER chaperone BiP. *Semin Cell Dev Biol*. 1999;10:465–472.
- Simmen T, Lynes EM, Gesson K, Thomas G. Oxidative protein folding in the endoplasmic reticulum: tight links to the mitochondria-associated membrane (MAM). *Biochim Biophys Acta*. 2010;1798:1465–73.
- Malhotra JD, Miao H, Zhang K, Wolfson A, Pennathur S, Pipe SW, Kaufman RJ. Antioxidants reduce endoplasmic reticulum stress and improve protein secretion. *Proc Natl Acad Sci U S A*. 2008;105:18525–18530.
- Kaufman RJ. Orchestrating the unfolded protein response in health and disease. *J Clin Invest*. 2002;110:1389–1398.
- Bertolotti A, Zhang Y, Hendershot LM, Harding HP, Ron D. Dynamic interaction of BiP and ER stress transducers in the unfolded-protein response. *Nat Cell Biol*. 2000;2:326–332.
- Bernales Sn, Papa FR, Walter P. Intracellular signaling by the unfolded protein response. *Annu Rev Cell Dev Biol*. 2006;22:487–508.
- Sawada T, Minamino T, Fu HY, Asai M, Okuda K, Isomura T, Yamazaki S, Asano Y, Okada K, Tsukamoto O, Sanada S, Asanuma H, Asakura M, Takashima S, Kitakaze M, Komuro I. X-box binding protein 1 regulates brain natriuretic peptide through a novel AP1/CRE-like element in cardiomyocytes. *J Mol Cell Cardiol*. 2010;48:1280–1289.
- Park SW, Zhou Y, Lee J, Lu A, Sun C, Chung J, Ueki K, Ozcan U. The regulatory subunits of PI3K, p85alpha and p85beta, interact with XBP-1 and increase its nuclear translocation. *Nat Med*. 2010;16:429–437.
- Urano F, Wang X, Bertolotti A, Zhang Y, Chung P, Harding HP, Ron D. Coupling of stress in the ER to activation of JNK protein kinases by transmembrane protein kinase IRE1. *Science*. 2000;287:664–666.
- Nishitoh H, Matsuzawa A, Tobiume K, Saegusa K, Takeda K, Inoue K, Hori S, Kakizuka A, Ichijo H. ASK1 is essential for endoplasmic reticulum stress-induced neuronal cell death triggered by expanded polyglutamine repeats. *Genes Dev*. 2002;16:1345–1355.
- Nakagawa T, Zhu H, Morishima N, Li E, Xu J, Yankner BA, Yuan J. Caspase-12 mediates endoplasmic-reticulum-specific apoptosis and cytotoxicity by amyloid-beta. *Nature*. 2000;403:98–103.
- Saleh M, Mathison JC, Wolinski MK, Bensinger SJ, Fitzgerald P, Droin N, Ulevitch RJ, Green DR, Nicholson DW. Enhanced bacterial clearance and sepsis resistance in caspase-12-deficient mice. *Nature*. 2006;440:1064–1068.
- Yamaguchi O, Higuchi Y, Hirotsu S, Kashiwase K, Nakayama H, Hikoso S, Takeda T, Watanabe T, Asahi M, Taniike M, Matsumura Y, Tsujimoto I, Hongo K, Kusakari Y, Kurihara S, Nishida K, Ichijo H, Hori M, Otsu K. Targeted deletion of apoptosis signal-regulating kinase 1 attenuates left ventricular remodeling. *Proc Natl Acad Sci U S A*. 2003;100:15883–15888.
- Homma K, Katagiri K, Nishitoh H, Ichijo H. Targeting ASK1 in ER stress-related neurodegenerative diseases. *Expert Opin Ther Targets*. 2009;13:653–664.
- Kaneko M, Niinuma Y, Nomura Y. Activation signal of nuclear factor-kappa B in response to endoplasmic reticulum stress is transduced via IRE1 and tumor necrosis factor receptor-associated factor 2. *Biol Pharm Bull*. 2003;26:931–935.
- Deng J, Lu PD, Zhang Y, Scheuner D, Kaufman RJ, Sonenberg N, Harding HP, Ron D. Translational repression mediates activation of nuclear factor kappa B by phosphorylated translation initiation factor 2. *Mol Cell Biol*. 2004;24:10161–10168.
- Ma Y, Hendershot LM. Delineation of a negative feedback regulatory loop that controls protein translation during endoplasmic reticulum stress. *J Biol Chem*. 2003;278:34864–34873.
- Zinszner H, Kuroda M, Wang X, Batchvarova N, Lightfoot RT, Remotti H, Stevens JL, Ron D. CHOP is implicated in programmed cell death in response to impaired function of the endoplasmic reticulum. *Genes Dev*. 1998;12:982–995.
- Tajiri S, Oyadomari S, Yano S, Morioka M, Gotoh T, Hamada JI, Ushio Y, Mori M. Ischemia-induced neuronal cell death is mediated by the endoplasmic reticulum stress pathway involving CHOP. *Cell Death Differ*. 2004;11:403–415.
- Oyadomari S, Koizumi A, Takeda K, Gotoh T, Akira S, Araki E, Mori M. Targeted disruption of the Chop gene delays endoplasmic reticulum stress-mediated diabetes. *J Clin Invest*. 2002;109:525–532.
- Fu HY, Okada K, Liao Y, Tsukamoto O, Isomura T, Asai M, Sawada T, Okuda K, Asano Y, Sanada S, Asanuma H, Asakura M, Takashima S, Komuro I, Kitakaze M, Minamino T. Ablation of C/EBP homologous protein attenuates ER-mediated apoptosis and cardiac dysfunction induced by pressure overload. *Circulation*. 2010;122:361–369.
- Cheng WP, Wang BW, Shyu KG. Regulation of GADD153 induced by mechanical stress in cardiomyocytes. *Eur J Clin Invest*. 2009;39:960–971.
- McCullough KD, Martindale JL, Klotz LO, Aw TY, Holbrook NJ. Gadd153 sensitizes cells to endoplasmic reticulum stress by down-regulating Bcl2 and perturbing the cellular redox state. *Mol Cell Biol*. 2001;21:1249–1259.
- Puthalakath H, O'Reilly LA, Gunn P, Lee L, Kelly PN, Huntington ND, Hughes PD, Michalak EM, McKimm-Breschkin J, Motoyama N, Gotoh T, Akira S, Bouillet P, Strasser A. ER stress triggers apoptosis by activating BH3-only protein Bim. *Cell*. 2007;129:1337–1349.
- Omori Y, Imai J-i, Watanabe M, Komatsu T, Suzuki Y, Kataoka K, Watanabe S, Tanigami A, Sugano S. CREB-H: a novel mammalian transcription factor belonging to the CREB/ATF family and functioning via the box-B element with a liver-specific expression. *Nucl Acids Res*. 2001;29:2154–2162.
- Kondo S, Murakami T, Tatsumi K, Ogata M, Kanemoto S, Otori K, Iseki K, Wanaka A, Imaizumi K. OASIS, a CREB/ATF-family member, modulates UPR signalling in astrocytes. *Nat Cell Biol*. 2005;7:186–194.
- Isodono K, Takahashi T, Imoto H, Nakanishi N, Ogata T, Asada S, Adachi A, Ueyama T, Oh H, Matsubara H. PARM-1 is an endoplasmic reticulum molecule involved in endoplasmic reticulum stress-induced apoptosis in rat cardiac myocytes. *PLoS One*. 2010;5:e9746.
- Tsukamoto O, Minamino T, Kitakaze M. Functional alterations of cardiac proteasomes under physiological and pathological conditions. *Cardiovasc Res*. 2010;85:339–346.
- Su H, Wang X. The ubiquitin-proteasome system in cardiac proteinopathy: a quality control perspective. *Cardiovasc Res*. 2010;85:253–262.
- Glembotski CC. The role of the unfolded protein response in the heart. *J Mol Cell Cardiol*. 2008;44:453–459.
- Terai K, Hiramoto Y, Masaki M, Sugiyama S, Kuroda T, Hori M, Kawase I, Hirota H. AMP-activated protein kinase protects cardiomyocytes against hypoxic injury through attenuation of endoplasmic reticulum stress. *Mol Cell Biol*. 2005;25:9554–9575.
- Okada K, Minamino T, Tsukamoto Y, Liao Y, Tsukamoto O, Takashima S, Hirata A, Fujita M, Nagamachi Y, Nakatani T, Yutani C, Ozawa K, Ogawa S, Tomoike H, Hori M, Kitakaze M. Prolonged endoplasmic reticulum stress in hypertrophic and failing heart after aortic constriction: possible contribution of endoplasmic reticulum stress to cardiac myocyte apoptosis. *Circulation*. 2004;110:705–712.
- Zhao H, Liao Y, Minamino T, Asano Y, Asakura M, Kim J, Asanuma H, Takashima S, Hori M, Kitakaze M. Inhibition of cardiac remodeling by pravastatin is associated with amelioration of endoplasmic reticulum stress. *Hypertens Res*. 2008;31:1977–1987.

40. Lee do Y, Lee KS, Lee HJ, Kim do H, Noh YH, Yu K, Jung HY, Lee SH, Lee JY, Youn YC, Jeong Y, Kim DK, Lee WB, Kim SS. Activation of PERK signaling attenuates Abeta-mediated ER stress. *PLoS One*. 2010;5:e10489.
41. Pattison JS, Sanbe A, Maloyan A, Osinska H, Kleivitsky R, Robbins J. Cardiomyocyte expression of a polyglutamine preamyloid oligomer causes heart failure. *Circulation*. 2008;117:2743–2751.
42. Zhang K. Integration of ER stress, oxidative stress and the inflammatory response in health and disease. *Int J Clin Exp Med*. 2010;3:33–40.
43. Karaskov E, Scott C, Zhang L, Teodoro T, Ravazzola M, Volchuk A. Chronic palmitate but not oleate exposure induces endoplasmic reticulum stress, which may contribute to INS-1 pancreatic {beta}-cell apoptosis. *Endocrinology*. 2006;147:3398–3407.
44. Maron BJ, Ferrans VJ, Roberts WC. Ultrastructural features of degenerated cardiac muscle cells in patients with cardiac hypertrophy. *Am J Pathol*. 1975;79:387–434.
45. Dally S, Monceau V, Corvazier E, Bredoux R, Raies A, Bobe R, del Monte F, Enouf J. Compartmentalized expression of three novel sarco/endoplasmic reticulum Ca²⁺-ATPase 3 isoforms including the switch to ER stress, SERCA3f, in non-failing and failing human heart. *Cell Calcium*. 2009;45:144–154.
46. Tsukamoto O, Minamoto T, Okada K-i, Shintani Y, Takashima S, Kato H, Liao Y, Okazaki H, Asai M, Hirata A, Fujita M, Asano Y, Yamazaki S, Asanuma H, Hori M, Kitakaze M. Depression of proteasome activities during the progression of cardiac dysfunction in pressure-overloaded heart of mice. *Biochem Biophys Res Commun*. 2006;340:1125–1133.
47. Weekes J, Morrison K, Mullen A, Wait R, Barton P, Dunn MJ. Hyperubiquitination of proteins in dilated cardiomyopathy. *Proteomics*. 2003;3:208–216.
48. Hamada H, Suzuki M, Yuasa S, Mimura N, Shinozuka N, Takada Y, Nishino T, Nakaya H, Koseki H, Aoe T. Dilated cardiomyopathy caused by aberrant endoplasmic reticulum quality control in mutant KDEL receptor transgenic mice. *Mol Cell Biol*. 2004;24:8007–8017.
49. Terrell D, Robbins J. Protein conformation-based disease: getting to the heart of the matter. *Annu Rev Physiol*. 2010;72:15–17.
50. Turdi S, Guo R, Huff AF, Wolf EM, Culver B, Ren J. Cardiomyocyte contractile dysfunction in the APPsw/PS1dE9 mouse model of Alzheimer's disease. *PLoS One*. 2009;4:e6033.
51. Su H-L, Liao C-L, Lin Y-L. Japanese encephalitis virus infection initiates endoplasmic reticulum stress and an unfolded protein response. *J Virol*. 2002;76:4162–4171.
52. Jheng JR, Lau KS, Tang WF, Wu MS, Horng JT. Endoplasmic reticulum stress is induced and modulated by enterovirus 71. *Cell Microbiol*. 2010;12:796–813.
53. Mao W, Fukuoka S, Iwai C, Liu J, Sharma VK, Sheu SS, Fu M, Liang CS. Cardiomyocyte apoptosis in autoimmune cardiomyopathy: mediated via endoplasmic reticulum stress and exaggerated by norepinephrine. *Am J Physiol Heart Circ Physiol*. 2007;293:H1636–H1645.
54. Mao W, Iwai C, Liu J, Sheu SS, Fu M, Liang CS. Darbeopetin alfa exerts a cardioprotective effect in autoimmune cardiomyopathy via reduction of ER stress and activation of the PI3K/Akt and STAT3 pathways. *J Mol Cell Cardiol*. 2008;45:250–260.
55. Hausenloy DJ, Yellon DM. Preconditioning and postconditioning: United at reperfusion. *Pharmacol Ther*. 2007;116:173–191.
56. Thuerauf DJ, Marcinko M, Gude N, Rubio M, Sussman MA, Glembofski CC. Activation of the unfolded protein response in infarcted mouse heart and hypoxic cultured cardiac myocytes. *Circ Res*. 2006;99:275–282.
57. Severino A, Campioni M, Straino S, Salloum FN, Schmidt N, Herbrand U, Frede S, Toietta G, Di Rocco G, Bussani R, Silvestri F, Piro M, Liuzzo G, Biasucci LM, Mellone P, Feroce F, Capogrossi M, Baldi F, Fandrey J, Ehrmann M, Crea F, Abbate A, Baldi A. Identification of protein disulfide isomerase as a cardiomyocyte survival factor in ischemic cardiomyopathy. *J Am Coll Cardiol*. 2007;50:1029–1037.
58. Toko H, Takahashi H, Kayama Y, Okada S, Minamoto T, Terasaki F, Kitaura Y, Komuro I. ATF6 is important under both pathological and physiological states in the heart. *J Mol Cell Cardiol*. 2010;49:113–120.
59. Martindale JJ, Fernandez R, Thuerauf D, Whittaker R, Gude N, Sussman MA, Glembofski CC. Endoplasmic reticulum stress gene induction and protection from ischemia/reperfusion injury in the hearts of transgenic mice with a tamoxifen-regulated form of ATF6. *Circ Res*. 2006;98:1186–1193.
60. Tadimalla A, Belmont PJ, Thuerauf DJ, Glassy MS, Martindale JJ, Gude N, Sussman MA, Glembofski CC. Mesencephalic astrocyte-derived neurotrophic factor is an ischemia-inducible secreted endoplasmic reticulum stress response protein in the heart. *Circ Res*. 2008;103:1249–1258.
61. Belmont PJ, Chen WJ, San Pedro MN, Thuerauf DJ, Gellings Lowe N, Gude N, Hilton B, Wolkowicz R, Sussman MA, Glembofski CC. Roles for endoplasmic reticulum-associated degradation and the novel endoplasmic reticulum stress response gene Derlin-3 in the ischemic heart. *Circ Res*. 2010;106:307–316.
62. Vitadello M, Penzo D, Petronilli V, Michieli G, Gomirato S, Menabo R, Di Lisa F, Gorza L. Overexpression of the stress protein Grp94 reduces cardiomyocyte necrosis due to calcium overload and simulated ischemia. *FASEB J*. 2003;17:923–925.
63. Shintani-Ishida K, Nakajima M, Uemura K, Yoshida K. Ischemic preconditioning protects cardiomyocytes against ischemic injury by inducing GRP78. *Biochem Biophys Res Commun*. 2006;345:1600–1605.
64. Liu XH, Zhang ZY, Sun S, Wu XD. Ischemic postconditioning protects myocardium from ischemia/reperfusion injury through attenuating endoplasmic reticulum stress. *Shock*. 2008;30:422–427.
65. Nickson P, Toth A, Erhardt P. PUMA is critical for neonatal cardiomyocyte apoptosis induced by endoplasmic reticulum stress. *Cardiovasc Res*. 2007;73:48–56.
66. Toth A, Jeffers JR, Nickson P, Min JY, Morgan JP, Zambetti GP, Erhardt P. Targeted deletion of Puma attenuates cardiomyocyte death and improves cardiac function during ischemia-reperfusion. *Am J Physiol Heart Circ Physiol*. 2006;291:H52–H60.
67. Avery J, Etzion S, DeBosch BJ, Jin X, Lupu TS, Beitinjaneh B, Grand J, Kovacs A, Sambandam N, Muslin AJ. TRB3 function in cardiac endoplasmic reticulum stress. *Circ Res*. 2010;106:1516–1523.
68. Woo CW, Cui D, Arellano J, Dorweiler B, Harding H, Fitzgerald KA, Ron D, Tabas I. Adaptive suppression of the ATF4-CHOP branch of the unfolded protein response by toll-like receptor signalling. *Nat Cell Biol*. 2009;11:1473–1480.
69. Martinon F, Chen X, Lee AH, Glimcher LH. TLR activation of the transcription factor XBP1 regulates innate immune responses in macrophages. *Nat Immunol*. 2010;11:411–418.
70. Ha T, Hu Y, Liu L, Lu C, McMullen JR, Kelley J, Kao RL, Williams DL, Gao X, Li C. TLR2 ligands induce cardioprotection against ischemia/reperfusion injury through a PI3K/Akt-dependent mechanism. *Cardiovasc Res*. 2010;87:694–703.
71. Sakata Y, Dong J-W, Vallejo JG, Huang C-H, Baker JS, Tracey KJ, Tacheuchi O, Akira S, Mann DL. Toll-like receptor 2 modulates left ventricular function following ischemia-reperfusion injury. *Am J Physiol Heart Circ Physiol*. 2007;292:H503–H509.
72. Kerkela R, Grazette L, Yacobi R, Iliescu C, Patten R, Beahm C, Walters B, Shevtsov S, Pesant S, Clubb FJ, Rosenzweig A, Salomon RN, Van Etten RA, Alroy J, Durand JB, Force T. Cardiotoxicity of the cancer therapeutic agent imatinib mesylate. *Nat Med*. 2006;12:908–916.
73. Force T, Kerkela R. Cardiotoxicity of the new cancer therapeutics—mechanisms of, and approaches to, the problem. *Drug Discov Today*. 2008;13:778–784.
74. Enrico O, Gabriele B, Nadia C, Sara G, Daniele V, Giulia C, Antonio S, Mario P. Unexpected cardiotoxicity in haematological bortezomib treated patients. *Br J Haematol*. 2007;138:396–397.
75. Fu HY, Minamoto T, Tsukamoto O, Sawada T, Asai M, Kato H, Asano Y, Fujita M, Takashima S, Hori M, Kitakaze M. Overexpression of endoplasmic reticulum-resident chaperone attenuates cardiomyocyte death induced by proteasome inhibition. *Cardiovasc Res*. 2008;79:600–610.
76. Kudo T, Kanemoto S, Hara H, Morimoto N, Morihara T, Kimura R, Tabira T, Imaizumi K, Takeda M. A molecular chaperone inducer protects neurons from ER stress. *Cell Death Differ*. 2008;15:364–375.
77. Wang G, Yang ZQ, Zhang K. Endoplasmic reticulum stress response in cancer: molecular mechanism and therapeutic potential. *Am J Transl Res*. 2010;2:65–74.
78. Healy SJ, Gorman AM, Mousavi-Shafaei P, Gupta S, Samali A. Targeting the endoplasmic reticulum-stress response as an anticancer strategy. *Eur J Pharmacol*. 2009;625:234–246.
79. Hansson GK. Inflammation, atherosclerosis, and coronary artery disease. *N Engl J Med*. 2005;352:1685–1695.
80. Feaver RE, Hastings NE, Pryor A, Blackman BR. GRP78 Upregulation by atheroprone shear stress Via p38-, {alpha}2{beta}1-dependent mechanism in endothelial cells. *Arterioscler Thromb Vasc Biol*. 2008;28:1534–1541.
81. Civelek M, Manduchi E, Riley RJ, Stoeckert CJ Jr, Davies PF. Chronic endoplasmic reticulum stress activates unfolded protein response in

- arterial endothelium in regions of susceptibility to atherosclerosis. *Circ Res*. 2009;105:453–461.
82. Dong Y, Zhang M, Liang B, Xie Z, Zhao Z, Asfa S, Choi HC, Zou MH. Reduction of AMP-activated protein kinase $\alpha 2$ increases endoplasmic reticulum stress and atherosclerosis in vivo. *Circulation*. 2010;121:792–803.
 83. Tabas I, Seimon T, Timmins J, Li G, Lim W. Macrophage apoptosis in advanced atherosclerosis. *Ann N Y Acad Sci*. 2009;1173(Suppl 1):E40–E45.
 84. Zhou J, Lhotak S, Hilditch BA, Austin RC. Activation of the unfolded protein response occurs at all stages of atherosclerotic lesion development in apolipoprotein E-deficient mice. *Circulation*. 2005;111:1814–1821.
 85. Feng B, Yao PM, Li Y, Devlin CM, Zhang D, Harding HP, Sweeney M, Rong JX, Kuriakose G, Fisher EA, Marks AR, Ron D, Tabas I. The endoplasmic reticulum is the site of cholesterol-induced cytotoxicity in macrophages. *Nat Cell Biol*. 2003;5:781–792.
 86. DeVries-Seimon T, Li Y, Yao PM, Stone E, Wang Y, Davis RJ, Flavell R, Tabas I. Cholesterol-induced macrophage apoptosis requires ER stress pathways and engagement of the type A scavenger receptor. *J Cell Biol*. 2005;171:61–73.
 87. Uysal KT, Scheja L, Wiesbrock SM, Bonner-Weir S, Hotamisligil GS. Improved glucose and lipid metabolism in genetically obese mice lacking $\alpha 2$. *Endocrinology*. 2000;141:3388–3396.
 88. Erbay E, Babaev VR, Mayers JR, Makowski L, Charles KN, Snitow ME, Fazio S, Wiest MM, Watkins SM, Linton MF, Hotamisligil GS. Reducing endoplasmic reticulum stress through a macrophage lipid chaperone alleviates atherosclerosis. *Nat Med*. 2009;15:1383–1391.
 89. Van Herck JL, De Meyer GR, Martinet W, Bult H, Vrints CJ, Herman AG. Proteasome inhibitor bortezomib promotes a rupture-prone plaque phenotype in ApoE-deficient mice. *Basic Res Cardiol*. 2010;105:39–50.
 90. Myoishi M, Hao H, Minamino T, Watanabe K, Nishihira K, Hatakeyama K, Asada Y, Okada K, Ishibashi-Ueda H, Gabbiani G, Bochaton-Piallat ML, Mochizuki N, Kitakaze M. Increased endoplasmic reticulum stress in atherosclerotic plaques associated with acute coronary syndrome. *Circulation*. 2007;116:1226–1233.
 91. Thorp E, Li G, Seimon TA, Kuriakose G, Ron D, Tabas I. Reduced apoptosis and plaque necrosis in advanced atherosclerotic lesions of ApoE $^{-/-}$ and Ldlr $^{-/-}$ mice lacking CHOP. *Cell Metab*. 2009;9:474–481.
 92. Doroudgar S, Thuerauf DJ, Marcinko MC, Belmont PJ, Glembotski CC. Ischemia activates the ATF6 branch of the endoplasmic reticulum stress response. *J Biol Chem*. 2009;284:29735–29745.
 93. Boyce M, Py BF, Ryazanov AG, Minden JS, Long K, Ma D, Yuan J. A pharmacoproteomic approach implicates eukaryotic elongation factor 2 kinase in ER stress-induced cell death. *Cell Death Differ*. 2008;15:589–599.
 94. Sokka A-L, Putkonen N, Mudo G, Pryazhnikov E, Reijonen S, Khiroug L, Belluardo N, Lindholm D, Korhonen L. Endoplasmic reticulum stress inhibition protects against excitotoxic neuronal injury in the rat brain. *J Neurosci*. 2007;27:901–908.
 95. Cnop M, Ladrerie L, Hekerman P, Ortis F, Cardozo AK, Dogusan Z, Flamez D, Boyce M, Yuan J, Eizirik DL. Selective inhibition of eukaryotic translation initiation factor 2 α dephosphorylation potentiates fatty acid-induced endoplasmic reticulum stress and causes pancreatic beta-Cell dysfunction and apoptosis. *J Biol Chem*. 2007;282:3989–3997.
 96. Korenykh AV, Egea PF, Korostelev AA, Finer-Moore J, Zhang C, Shokat KM, Stroud RM, Walter P. The unfolded protein response signals through high-order assembly of Ire1. *Nature*. 2009;457:687–693.
 97. Di Lorenzo G, Autorino R, Bruni G, Carteni G, Ricevuto E, Tudini M, Ficorella C, Romano C, Aieta M, Giordano A, Giuliano M, Gonnella A, De Nunzio C, Rizzo M, Montesarchio V, Ewer M, De Placido S. Cardiovascular toxicity following sunitinib therapy in metastatic renal cell carcinoma: a multicenter analysis. *Ann Oncol*. 2009;20:1535–1542.
 98. Chu TF, Rupnick MA, Kerkela R, Dallabrida SM, Zurakowski D, Nguyen L, Woulfe K, Pravda E, Cassiola F, Desai J, George S, Morgan JA, Harris DM, Ismail NS, Chen J-H, Schoen FJ, Van den Abbeele AD, Demetri GD, Force T, Chen MH. Cardiotoxicity associated with tyrosine kinase inhibitor sunitinib. *Lancet*. 2007;370:2011–2019.
 99. Takahama H, Minamino T, Asanuma H, Fujita M, Asai T, Wakeno M, Sasaki H, Kikuchi H, Hashimoto K, Oku N, Asakura M, Kim J, Takashima S, Komamura K, Sugimachi M, Mochizuki N, Kitakaze M. Prolonged targeting of ischemic/reperfused myocardium by liposomal adenosine augments cardioprotection in rats. *J Am Coll Cardiol*. 2009;53:709–717.
 100. Gross E, Sevier CS, Heldman N, Vitu E, Bentzur M, Kaiser CA, Thorpe C, Fass D. Generating disulfides enzymatically: reaction products and electron acceptors of the endoplasmic reticulum thiol oxidase Ero1p. *Proc Natl Acad Sci U S A*. 2006;103:299–304.
 101. Austin RC. The unfolded protein response in health and disease. *Antioxid Redox Signal*. 2009;11:2279–2287.
 102. Blais JD, Chin K-T, Zito E, Zhang Y, Heldman N, Harding HP, Fass D, Thorpe C, Ron D. A Small molecule inhibitor of endoplasmic reticulum oxidation 1 (ERO1) with selectively reversible thiol reactivity. *J Biol Chem*. 2010;285:20993–21003.
 103. Guo R, Ma H, Gao F, Zhong L, Ren J. Metallothionein alleviates oxidative stress-induced endoplasmic reticulum stress and myocardial dysfunction. *J Mol Cell Cardiol*. 2009;47:228–237.
 104. Pal R, Cristan EA, Schnittker K, Narayan M. Rescue of ER oxidoreductase function through polyphenolic phytochemical intervention: implications for subcellular traffic and neurodegenerative disorders. *Biochem Biophys Res Commun*. 2010;392:567–571.
 105. Wang X-Z, Ron D. Stress-induced phosphorylation and activation of the transcription factor CHOP (GADD153) by p38 MAP kinase. *Science*. 1996;272:1347–1349.
 106. Hikoso S, Ikeda Y, Yamaguchi O, Takeda T, Higuchi Y, Hirohata S, Kashiwase K, Yamada M, Asahi M, Matsumura Y, Nishida K, Matsuzaki M, Hori M, Otsu K. Progression of heart failure was suppressed by inhibition of apoptosis signal-regulating kinase 1 via transcortin gene transfer. *J Am Coll Cardiol*. 2007;50:453–462.
 107. Asai M, Tsukamoto O, Minamino T, Asanuma H, Fujita M, Asano Y, Takahama H, Sasaki H, Higo S, Asakura M, Takashima S, Hori M, Kitakaze M. PKA rapidly enhances proteasome assembly and activity in vivo canine hearts. *J Mol Cell Cardiol*. 2009;46:452–462.
 108. Prins D, Michalak M. Endoplasmic reticulum proteins in cardiac development and dysfunction. *Can J Physiol Pharmacol*. 2009;87:419–425.
 109. Cohen FE, Kelly JW. Therapeutic approaches to protein-misfolding diseases. *Nature*. 2003;426:905–909.
 110. Reddy RK, Mao C, Baumeister P, Austin RC, Kaufman RJ, Lee AS. Endoplasmic reticulum chaperone protein GRP78 protects cells from apoptosis induced by topoisomerase inhibitors: role of ATP binding site in suppression of caspase-7 activation. *J Biol Chem*. 2003;278:20915–20924.
 111. Ulloa-Aguirre A, Janovick JA, Brothers SP, Conn PM. Pharmacologic rescue of conformationally-defective proteins: implications for the treatment of human disease. *Traffic*. 2004;5:821–837.
 112. Perlmutter DH. Chemical chaperones: a pharmacological strategy for disorders of protein folding and trafficking. *Pediatr Res*. 2002;52:832–836.
 113. Leonard JV, Morris AAM. Urea cycle disorders. *Semin Neonatol*. 2002;7:27–35.
 114. Lee B, Rhead W, Diaz GA, Scharschmidt BF, Mian A, Shchelochkov O, Marier JF, Beliveau M, Mauney J, Dickinson K, Martinez A, Gargosky S, Mokhtarani M, Berry SA. Phase 2 comparison of a novel ammonia scavenging agent with sodium phenylbutyrate in patients with urea cycle disorders: safety, pharmacokinetics and ammonia control. *Mol Genet Metab*. 2010;100:221–228.
 115. Cudkovicz ME, Andres PL, Macdonald SA, Bedlack RS, Choudry R, Brown RH Jr, Zhang H, Schoenfeld DA, Shefner J, Matson S, Matson WR, Ferrante RJ, ALS TN, CONSORTIUMS TNVAR. Phase 2 study of sodium phenylbutyrate in ALS. *Amyotroph Lateral Scler*. 2009;10:99–106.
 116. Ozcan Y, Yilmaz E, Ozcan L, Furuhashi M, Vaillancourt E, Smith RO, Gorgun CZ, Hotamisligil GS. Chemical chaperones reduce ER stress and restore glucose homeostasis in a mouse model of type 2 diabetes. *Science*. 2006;313:1137–1140.
 117. Wiley JC, Meabon JS, Frankowski H, Smith EA, Schecterson LC, Bothwell M, Ladiges WC. Phenylbutyric acid rescues endoplasmic reticulum stress-induced suppression of APP proteolysis and prevents apoptosis in neuronal cells. *PLoS One*. 2010;5:e9135.
 118. Basseri S, Lhotak S, Sharma AM, Austin RC. The chemical chaperone 4-phenylbutyrate inhibits adipogenesis by modulating the unfolded protein response. *J Lipid Res*. 2009;50:2486–2501.
 119. Daosukho C, Chen Y, Noel T, Sompol P, Nithipongvanitch R, Velez JM, Oberley TD, St. Clair DK. Phenylbutyrate, a histone deacetylase

- inhibitor, protects against Adriamycin-induced cardiac injury. *Free Radic Biol Med*. 2007;42:1818–1825.
120. Yoshiuchi K, Kaneto H, Matsuoka TA, Kasami R, Kohno K, Iwawaki T, Nakatani Y, Yamasaki Y, Shimomura I, Matsuhisa M. Pioglitazone reduces ER stress in the liver: direct monitoring of in vivo ER stress using ER stress-activated indicator transgenic mice. *Endocr J*. 2009;56:1103–1111.
121. Sasaki H, Asanuma H, Fujita M, Takahama H, Wakeno M, Ito S, Ogai A, Asakura M, Kim J, Minamino T, Takashima S, Sanada S, Sugimachi M, Komamura K, Mochizuki N, Kitakaze M. Metformin prevents progression of heart failure in dogs: role of AMP-activated protein kinase. *Circulation*. 2009;119:2568–2577.
122. Dong Y, Zhang M, Wang S, Liang B, Zhao Z, Liu C, Wu M, Choi HC, Lyons TJ, Zou M-H. Activation of AMP-activated protein kinase inhibits oxidized LDL-triggered endoplasmic reticulum stress in vivo. *Diabetes*. 2010;59:1386–1396.
123. Yusta B, Baggio LL, Estall JL, Koehler JA, Holland DP, Li H, Pipeleers D, Ling Z, Drucker DJ. GLP-1 receptor activation improves β cell function and survival following induction of endoplasmic reticulum stress. *Cell Metab*. 2006;4:391–406.
124. Ban K, Noyan-Ashraf MH, Hoefler J, Bolz S-S, Drucker DJ, Husain M. Cardioprotective and vasodilatory actions of glucagon-like peptide 1 receptor are mediated through both glucagon-like peptide 1 receptor-dependent and -independent pathways. *Circulation*. 2008;117:2340–2350.
125. Blais JD, Chin KT, Zito E, Zhang Y, Heldman N, Harding HP, Fass D, Thorpe C, Ron D. A small molecule inhibitor of endoplasmic reticulum oxidation 1 (ERO1) with selectively reversible thiol reactivity. *J Biol Chem*. 2010;285:20993–1003.
126. Kim I, Shu CW, Xu W, Shiau CW, Grant D, Vasile S, Cosford ND, Reed JC. Chemical biology investigation of cell death pathways activated by endoplasmic reticulum stress reveals cytoprotective modulators of ASK1. *J Biol Chem*. 2009;284:1593–1603.

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