ER stress and the unfolded protein response in neurodegeneration

Claudio Hetz[1–3] and Smita Saxena[4]

Abstract | The clinical manifestation of neurodegenerative diseases is initiated by the selective alteration in the functionality of distinct neuronal populations. The pathology of many neurodegenerative diseases includes accumulation of misfolded proteins in the brain. In physiological conditions, the proteostasis network maintains normal protein folding, trafficking and degradation; alterations in this network — particularly disturbances to the function of endoplasmic reticulum (ER) — are thought to contribute to abnormal protein aggregation. ER stress triggers a signalling reaction known as the unfolded protein response (UPR), which induces adaptive programmes that improve protein folding and promote quality control mechanisms and degradative pathways or can activate apoptosis when damage is irreversible. In this Review, we discuss the latest advances in defining the functional contribution of ER stress to brain diseases, including novel evidence that relates the UPR to synaptic function, which has implications for cognition and memory. A complex concept is emerging wherein the consequences of ER stress can differ drastically depending on the disease context and the UPR signalling pathway that is altered. Strategies to target specific components of the UPR using small molecules and gene therapy are in development, and promise interesting avenues for future interventions to delay or stop neurodegeneration.

[1]*Biomedical Neuroscience Institute, Faculty of Medicine, University of Chile, Independencia 1027, Santiago, Chile.*
[2]*Geroscience Center for Brain Health and Metabolism, Santiago, Chile.*
[3]*Program of Cellular and Molecular Biology, Institute of Biomedical Sciences, Center for Molecular Studies of the Cell, University of Chile, Independencia 1027, Santiago, Chile.*
[4]*Department of Neurology, Inselspital University Hospital, University of Bern, Freiburgstrasse 16, CH-3010 Bern, Switzerland.*
chetz@med.uchile.cl; smita.saxena@insel.ch

doi:10.1038/nrneurol.2017.99
Published online 21 Jul 2017

Neurodegenerative diseases are characterized by progressive loss of neuronal function in defined regions of the nervous system, culminating in severe dysfunction. These diseases include Alzheimer disease (AD), Parkinson disease (PD), amyotrophic lateral sclerosis (ALS), Huntington disease (HD), prion-related disorders, retinitis pigmentosa and some myelin-related disorders. Each of these conditions has distinct pathophysiological and clinical hallmarks, yet they share a pathological trait: abnormal aggregation of misfolded proteins[1–3] (BOX 1; FIG. 1). The causal association between accumulation of a specific misfolded protein and the development of pathology in these diseases means that they are often described as proteinopathies, or protein misfolding disorders (PMDs)[1,4].

Under physiological conditions, chaperones resident in the cytosol and the endoplasmic reticulum (ER) ensure precise folding of newly synthesized native proteins, and quality control mechanisms identify misfolded proteins and facilitate their degradation via the proteasome, lysosome and autophagy pathways[5]. This process, known as protein homeostasis or proteostasis[6], is fundamental to the maintenance of cellular health and function, as it prevents abnormal protein aggregation. However, sustaining cellular proteostasis becomes challenging and complex in PMDs in which misfolded proteins accumulate[7,8]. One consequence of accumulating misfolded protein is the generation of ER stress[9,10], which triggers a rapid and coordinated biochemical response that involves adaptive signalling pathways; this reaction is known as the unfolded protein response (UPR) (FIG. 2). Emerging evidence indicates that ER stress has a vital role in the pathophysiology of PMDs, although the organization, functions and regulation of ER folding and quality control mechanisms are not yet completely understood.

In this Review, we discuss the most recent advances in our understanding of the functional link between ER stress, the UPR, and neurodegeneration, not only in typical PMDs, but also in inflammatory disease and traumatic injury to the nervous system. We provide an in-depth mechanistic explanation of how disease-specific proteins affect ER proteostasis, and discuss novel evidence that links the physiological activity of the UPR with neuronal plasticity and synaptic function. Overall, we bring together emerging evidence that UPR activation occurs in human brain tissue, and analyse evidence from preclinical models that supports functional involvement of ER stress in neurological disease. We also discuss the latest efforts to develop therapeutic strategies for targeting the UPR in neurodegenerative diseases.

Key points

- Many neurodegenerative diseases involve the accumulation of protein aggregates
- Endoplasmic reticulum (ER) stress triggers activation of the unfolded protein response (UPR), an adaptive reaction that restores cellular protein homeostasis, known as proteostasis
- Dysfunction of proteostasis is associated with abnormal levels of ER stress and is associated with neuronal degeneration in human post-mortem brain tissue
- Targeting the UPR can have distinct and even opposite effects on disease progression, depending on the disease context and the signalling branch that is analysed
- Gene therapy and pharmacological strategies to attenuate ER stress alleviates degeneration in various disease models
- Chronic ER stress not only results in neuronal loss, but also represses the synthesis of synaptic proteins, with implications for cognition and memory, and possibly autism spectrum disorder

Molecular players in UPR signalling

A small fraction of PMDs are familial and involve a genetic mutation that causes misfolding of a specific protein, but in most sporadic cases of the diseases, the same proteins often accumulate and aggregate. This observation has led to the hypothesis that alterations in the buffering capacity of the proteostasis network contribute to the aetiology of PMDs. Importantly, impairment of proteostasis is also exacerbated during ageing[5,11], the main risk factor for PMDs (BOX 2).

The function of the ER and the secretory pathway are key elements of the proteostasis network that are altered in brain diseases. The ER is the site of folding for at least one-third of the proteome, a process that is assisted by a complex family of chaperones, foldases, cofactors and enzymes that all mediate various post-translational modifications. Several physiological and pathological conditions that alter the function of the secretory pathway result in ER stress; this perturbation activates the UPR, which controls the stability of RNAs and the rate of protein synthesis, and activates transcription of a large spectrum of genes involved in almost every aspect of the secretory pathway. Typical UPR target genes encode for proteins involved in protein folding, ER-associated degradation (ERAD), vesicular trafficking, autophagy, ER redox control, amino acid metabolism and lipid synthesis[9]. The most studied physiological role of the UPR in tissue homeostasis is its involvement in sustaining the function of specialized secretory cells, in which the pathway is particularly important owing to the high demand in these cells for protein synthesis; this activity creates physiological, rather than pathological, ER stress. Examples of such cells are B cells, endocrine and exocrine pancreatic cells and salivary gland cells[12].

The UPR can be viewed as a simple signal transduction pathway that involves two main components: stress sensors at the ER membrane, and downstream transcription factors that reprogramme gene expression toward stress mitigation or the induction of proapoptotic programmes[13] (FIG. 3). The UPR target genes vary depending on the tissue context and the type of physiological perturbation that causes ER stress. This variation might result from the formation of different heterodimers between transcription factors, post-translational modifications and epigenetic changes[14]. In mouse and human cells, differences in the patterns of gene expression triggered by ER stress have also been attributed to differences in genetic background[13,15]. Nevertheless, three main type-I transmembrane proteins initiate the UPR: inositol-requiring protein 1α (IRE1α; a β isoform is selectively expressed in the lung and intestine), activating transcription factor-6 (ATF6, α and β), and protein kinase RNA-like ER kinase (PERK).

Inositol-requiring protein 1

IRE1α is the most evolutionarily conserved ER stress transducer, and contains RNase and kinase domains within its cytosolic region. Upon activation, IRE1α dimerizes and autotransphosphorylates, leading to IRE1α-mediated removal of a 26-nucleotide intron from the mRNA that encodes the transcription factor XBP1 (REF. 16). The consequent shift in reading frame of the XBP1 mRNA results in expression of an active and stable transcription factor called XBP1s. The activity of XBP1s has been linked to prosurvival events that support proteostasis by inducing the expression of various genes that are involved in protein folding and quality-control mechanisms, and by activating the ERAD[14] (FIG. 3). XBP1s also increases biogenesis of the ER and Golgi compartments, thereby increasing the rate of protein secretion. In addition, IRE1α targets a group of mRNAs and microRNAs (possibly dependent on sequence, secondary structure and/or tissue location) for degradation through a process known as regulated IRE1α-dependent decay (RIDD)[17]. RIDD is thought to have pleiotropic effects in cells, as the phenomenon alters expression of multiple proteins with various functions; these effects include stress mitigation, inflammation and apoptosis[17]. Furthermore, active IRE1α can recruit several adaptor proteins that mediate crosstalk with other stress pathways, including the mitogen-activated protein (MAP) kinase pathway, autophagy, and inflammatory pathways that involve nuclear factor-κB[14]. IRE1α, therefore, has a dual role in the response to chronic ER stress, mediating adaptation through XBP1s and mediating induction of apoptosis via the MAP kinase pathway and RIDD. On this basis, small molecules that inhibit IRE1α signalling have been developed[18] and tested in the context of cancer, diabetes mellitus and retinal damage[19], and offer an interesting avenue of intervention in PMDs.

Activating transcription factor-6

Two isoforms of ATF6 — α and β — are expressed ubiquitously in the ER and contain a basic leucine zipper transcription factor domain in its cytosolic domain. ER stress causes ATF6 to translocate to the Golgi apparatus, where endopeptidase S1P and endopeptidase S2P cleave it and release a fragment called ATF6f that includes the transcription factor domain. ATF6f translocates to the nucleus and induces expression of XBP1 and genes that are involved in ERAD and protein folding[16] (FIG. 3). ATF6f cooperates with XBP1s to modulate gene expression under stress.

Autophagy
Self-degradation process with functions that include the removal of misfolded or aggregated proteins and damaged organelles.

Proteostasis
A portmanteau of the words protein and homeostasis, referring to the function of integrated biological pathways within cells that control the biogenesis, folding, trafficking and degradation of proteins present within and outside the cell.

ER stress
A cellular condition that involves accumulation of misfolded and/or unfolded proteins at the ER; ER stress activates the unfolded protein response, which enables adaptation to stress or triggers apoptosis of irreversibly-damaged cells.

Unfolded protein response
A signal transduction pathway that is activated by an accumulation of unfolded or misfolded proteins in the ER lumen; the unfolded protein response mediates adaptation to protein folding stress or the elimination of non-functional cells by apoptosis.

ER-associated degradation
Cellular pathway that targets misfolded proteins at the ER for ubiquitylation and subsequent degradation in the cytosol by the proteasome.

Integrated stress response
An adaptive pathway in eukaryotic cells that is activated by a range of stress conditions that converge on phosphorylation of eukaryotic translation initiation factor 2α, which leads to a decrease in global protein synthesis and the upregulation of selected genes that promote cellular homeostasis.

Protein disulfide isomerase
One of a family of enzymes in the ER that catalyse the formation, isomerization and breakage of disulfide bonds between cysteine residues within proteins as they fold, enabling the correct arrangement of disulfide bonds in the fully folded state to form quickly.

Protein kinase RNA-like ER kinase

PERK is emerging as a therapeutic target in neurodegenerative diseases, as its signalling pathway is involved in the control of protein synthesis. ER stress triggers the activation of PERK, which directly phosphorylates the protein translation initiation factor eIF2α, thereby inhibiting protein synthesis and consequently preventing an overload of proteins in the ER lumen[16]. In addition, phosphorylation of eIF2α leads to selective translation of the transcription factor ATF4, which is crucial for the upregulation of genes that encode proteins involved in redox control, amino acid metabolism, autophagy and protein folding and synthesis (FIG. 3). In cells with irreversible damage, ATF4 also engages cell death pathways via induction of the transcription factor DDIT-3 (also known as GADD153, and commonly known as CHOP), reactive oxygen species and members of the apoptosis regulator BCL-2 family[14]. The levels of eIF2α phosphorylation are controlled by serine/threonine-protein phosphatase PP1 (PP1) in complex with either a constitutive regulatory subunit called PPP1R15B or an ER-stress-induced form of the regulatory subunit called GADD34 (also known as PPP1R15A); expression of GADD34 is induced by ER-stress because its encoding gene is activated by CHOP[16]. Beyond the UPR, eIF2α phosphorylation is also a convergent point of several pathways that make up the so-called integrated stress response, which is triggered by various stimuli, including viral infection, nutrient starvation and haem deficiency[20].

Box 1 | **Protein misfolding associated with neurodegenerative diseases**

Alzheimer disease
- Deposits of intracellular tau aggregate to form neurofibrillary tangles.
- Extracellular aggregates of amyloid-β form amyloid plaques.

Parkinson disease
- Formation of protein inclusion bodies, called Lewy bodies, that contain aggregated α-synuclein and ubiquitin.
- Accumulation of tau deposits.

Huntington disease
- Expansion of the polyglutamine tract in the huntingtin protein causes intracellular aggregation of the protein.

Prion-related disorders (transmissible spongiform encephalopathies)
- Extracellular accumulation of the scrapie form of the prion protein, a pathological isoform of the normal cellular prion protein.

Amyotrophic lateral sclerosis
- Formation of protein inclusions in motor neurons, primarily composed of TDP-43.
- Repeat expansion in *C9orf72* leads to production of dipeptide repeat proteins such as glycine–alanine, which form inclusion bodies.
- Accumulation of mutant superoxide dismutase 1 and TAR DNA binding protein 43 (TDP-43).

Demyelinating disorders
- Accumulation of mutant myelin proteins at the endoplasmic reticulum in Schwann cells and oligodendrocytes.

Frontotemporal dementia
- Accumulation of repeat-associated non-ATG translation peptide products, TDP-43 and tau inclusions.

Retinitis pigmentosa
- Accumulation of misfolded mutant rhodopsin inside the endoplasmic reticulum of retinal cells.

Molecular triggers of ER stress

The specific mechanisms that underlie impairment of proteostasis in PMDs have started to be defined by studies that highlight distinct points in the secretory pathway that are disrupted (FIG. 4). For example, in AD, overexpression of β-amyloid precursor in neuronal cell cultures sensitizes cells to ER stress[21], and many studies (reviewed in REF. 22) have indicated that amyloid-β oligomers, which can accumulate in the ER lumen[23,24], cause disruption of ER calcium homeostasis, resulting in a proapoptotic ER stress response. A similar model has been proposed for prion-related disorders[25–27]. At the molecular level, reduced steady-state levels of calcium in the ER lumen results in suboptimal functioning of calcium-binding chaperones, such as calreticulin, endoplasmin (commonly known as GRP94), GRP78 (commonly known as BiP) and protein disulfide isomerase PDI (also known as PDIA1), leading to ER stress[28]. Current evidence suggests that AD-associated mutations in presenilin-1 and presenilin-2 alter ER calcium homeostasis in the same way, but might also reduce the activity of IRE1α and expression of BiP, thereby interfering with UPR signalling[29,30], although these results are under debate because some studies have indicated that presenilins do not affect ER stress[31,32]. Accumulation of tau triggers abnormal interactions between ER proteins[33] and essential components of ERAD, thereby impairing this pathway[34].

Several molecular mechanisms of proteostasis disruption have been identified in PD. Aggregates of α-synuclein tend to accumulate in the ER lumen and induce ER stress, possibly through an abnormal association with ER chaperones[35]. Notably, α-synuclein inhibits trafficking of proteins from the ER to the Golgi apparatus, thereby affecting protein maturation and consequently causing ER stress[36]. Direct impairment of the UPR by α-synuclein has also been proposed, via inhibition of ATF6 activation that results from a physical interaction[37]. PD-associated mutations in the RING-finger-containing E3 ubiquitin ligase parkin also causes ER stress that results in neuronal degeneration, possibly owing to altered proteasome-mediated degradation[38,39]. Finally, mutation of the PD-associated gene *ATP13A2* (also known as *PARK9*), which encodes cation-transporting ATPase 13A2, results in misfolding and accumulation of the protein at the ER lumen, triggering chronic activation of the UPR[40].

In ALS, several independent mechanisms are thought to adversely affect ER physiology. Mutant superoxide dismutase 1 (SOD1, encoded by *SOD1)*, physically interacts and inhibits a component of the ERAD pathway known as Derlin-1 (REFS 41,42). Mutant SOD1 has also been observed inside the ER lumen, where it might sequester vital chaperones, such as BiP and PDI[41,43,44]. Translocation of ALS-associated mutant FUS protein from the nucleus to the cytoplasm in motor neurons is

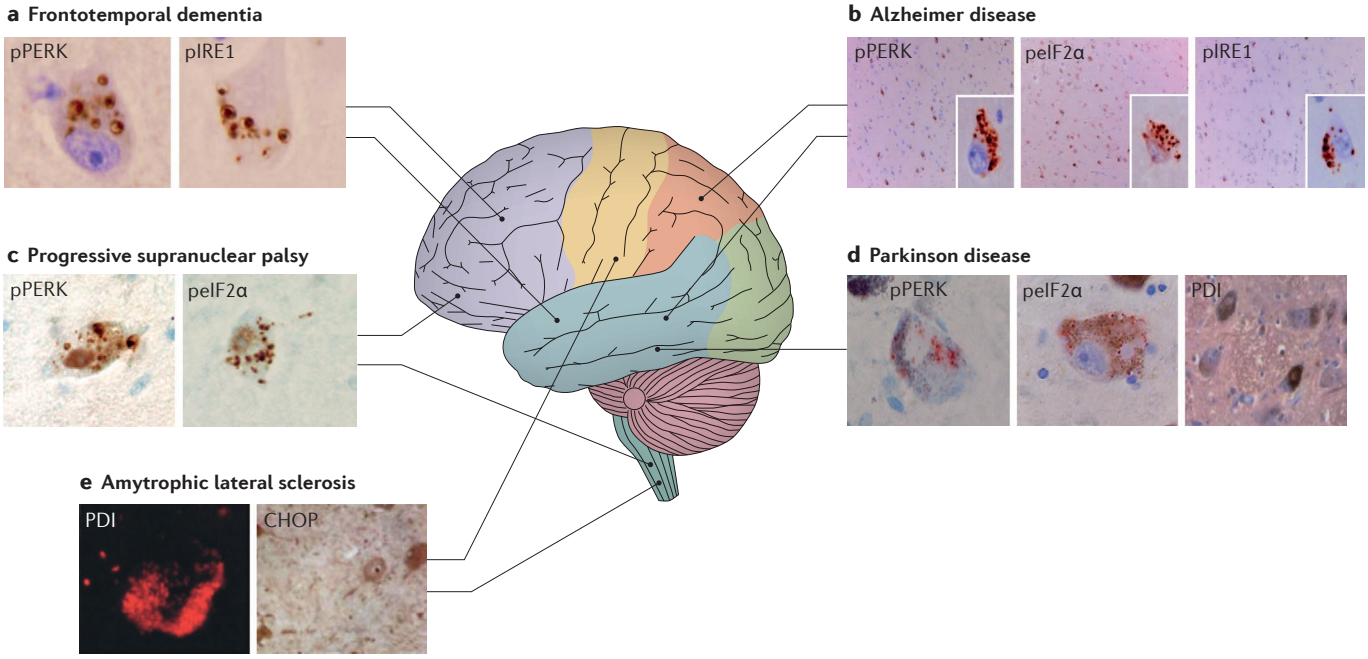

Figure 1 | **Protein aggregates in tissue from patients with neurodegenerative disease.** Most neurodegenerative diseases have distinct clinical manifestations, but they share accumulation of protein aggregates and inclusions that contain specific proteins in distinct brain regions; these aggregates are associated with an endoplasmic reticulum stress reaction. PDI, protein disulfide isomerase; p, phosphorylated; PERK, protein kinase RNA-like ER kinase; IRE1, inositol-requiring protein 1. Part **a** reproduced with permission from John Wiley and Sons © Nijholt, D. A. *et al. J. Pathol.* **226**, 693–702 (2012). Part **b** reproduced with permission from Elsevier © Jeroen, J. M. *et al. Am. J. Pathol.* **174**, 1241–1251 (2009). Part **c** reproduced with permission from BioMed Central © Stutzbach, L. D. *et al. Acta Neuropathol. Commun.* **1**, 31 (2013). Part **d** reproduced with permission from Elsevier © Hoozemans, J. J. *et al. Biochem. Biophys. Res. Commun.* **354**, 707–711 (2007) (left and middle panels) and © Conn, K. J. *et al. Brain Res.* **1022**, 164–172 (2004) (right panel). Part **e** reproduced with permission from Elsevier © Atkin, J. D. *et al. Neurobiol. Dis.* **30**, 400–407 (2008) (left panel) and © Ito, Y. *et al. Neurobiol. Dis.* **36**, 470–476 (2009) (right panel).

associated with ER stress and fragmentation of the Golgi apparatus[45]. FUS also interacts with PDI upon induction of ER stress[46], and the function of PDI is also affected by nitrosylation of the active site, which has been reported in ALS[47], PD and AD[48]. ALS-linked mutant vesicle-associated protein-associated protein B (VAPB) can interact with ATF6 and XBP1, altering their activities and subcellular distribution[49]. In patients with a *C9orf72* repeat expansion, the glycine–alanine dipeptide repeat protein causes neurotoxicity by initiating ER stress and altering the ubiquitin-proteasome system through an unknown mechanism[50].

Two studies have shown that mutant huntingtin impairs ERAD through abnormal protein–protein interactions[51,52]. In addition, fragments of huntingtin other than the polyglutamine tract can alter the morphology of the ER through an aberrant interaction with dynamin-1, resulting in ER dilatation and chronic stress[53]. Perturbations to many other molecular components of the secretory pathway have been associated with ER stress in PMDs[7,33,54]. Overall, the common alterations to ER proteostasis that are observed in neurodegenerative diseases highlight disruption of ER calcium homeostasis, vesicular trafficking, ERAD activity and UPR dysregulation, and abnormal ER chaperone function.

ER stress in human neurodegeneration

Most studies of the UPR in humans have been performed with autopsy brain samples (FIG. 1). Several neurodegenerative disorders have also been modelled with human induced pluripotent stem cells (iPSCs). Patient-specific iPSC-derived neuronal cells largely recapitulate relevant disease phenotypes, enabling the characterization of disease mechanisms and the screening for novel therapeutic targets. Across various PMDs, multiple studies of human tissue and iPSCs have demonstrated activation of the UPR. This consistency suggests that ER stress has a central and conserved role in the pathogenic neuronal response.

Studies of human tissue

In brain tissue from patients with AD, increases in UPR activation markers are widespread and occur early relative to markers in control brain tissue from people without dementia. For example, increased expression levels of the ER chaperone BiP) have also been observed in regions affected by AD, such as the hippocampus and temporal cortex[55,56]. Furthermore, augmented PERK and IRE1α signalling has been observed in AD neurons that contain abnormally phosphorylated tau[55–58]. Remarkably, levels of IRE1α phosphorylation directly correlated with the Braak stage of pathology in patients with AD[59].

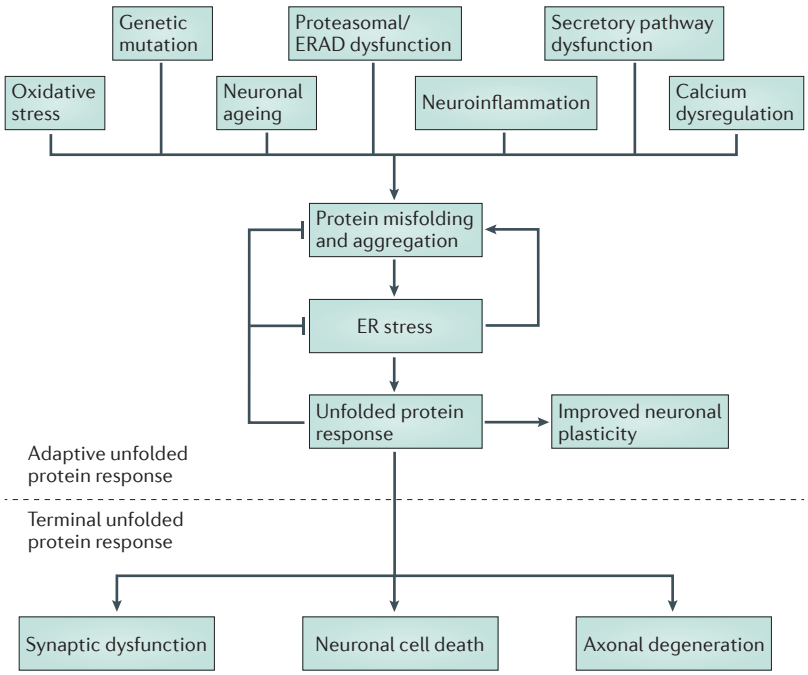

Figure 2 | **ER stress and proteostasis in neurodegenerative diseases.** Ageing, environmental factors and mutation of specific disease-related genes can trigger misfolding of a particular protein, leading to formation of aggregates that range from small oligomeric species to large inclusion bodies. This abnormal aggregation results in endoplasmic reticulum (ER) stress. ER stress can increase aggregation of disease-related proteins via a feedback loop by altering the folding and quality-control capacity of the cell or by altering the expression of disease-related genes. ER stress engages unfolded protein response (UPR) sensors that activate distinct downstream responses to improve protein folding and quality-control mechanisms to reduce ER stress. Long-term ER stress over-rides the adaptive responses of the UPR, and induces apoptosis. Chronic inhibition of protein synthesis can also reduce synthesis of synaptic proteins, thereby impairing neuronal function. ERAD, ER-associated degradation.

Increased UPR activation has also been reported in other tauopathies, such as progressive supranuclear palsy, Pick disease, familial FTD with parkinsonism linked to chromosome 17 (REF. 60), and frontotemporal lobar degeneration with tau pathology[60]. In PD, UPR activation has been observed early in the disease, associated with increased phosphorylation of PERK and eIF2α in neuromelanin-positive neurons of the substantia nigra[61,62]. Similarly, increased levels of UPR markers have been observed in multiple system atrophy, a disease that, like PD, is associated with accumulation of α-synuclein[63].

In ALS, expression of components of all three UPR pathways is increased in the spinal cord of patients with sporadic and familial forms of the disease[64–68]. Moreover, PDI is upregulated in the cerebrospinal fluid of patients with sporadic ALS, and its expression is induced in motor neurons of these patients, indicating that PDI dysfunction contributes to the ALS pathology and might aid diagnosis[64]. Proteomic screening for biomarkers in the blood of patients with sporadic ALS also identified that the major proteins upregulated were a cluster of ER-stress-induced chaperones[69]. Global gene expression profiling of brain tissue from patients with ALS associated with a *C9orf72* repeat expansion revealed that UPR alterations are a major signature of the pathology in cerebellum[70].

In prion-related disorders, such as Creutzfeldt–Jakob disease (CJD), elevated levels of ER stress markers, including protein disulfide isomerase A3 (commonly known as ERp57 and also known as GRP58), BiP and GRP94, have been observed in cortical samples from patients with sporadic forms of CJD and in those with infectious forms of the disease[25]. The main upregulated protein that was identified in a proteomic screening study in patients with CJD was ERp57 (REF. 71). However, monitoring of PERK and IRE1α phosphorylation in other studies failed to detect signs of ER stress in CJD[58,72].

In post-mortem cortex tissue from patients who had HD, increased transcript levels of the UPR effectors BiP, CHOP and the ERAD component HERP have been observed[73]. Analysis of global gene expression data has also revealed major alterations in UPR signalling in HD[74], and increased splicing of XBP1 has been observed in the striatum of patients with HD[75].

Studies of iPSC-derived human neurons

Analysis of cortical neurons that were generated from iPSCs from patients with PD caused by α-synuclein mutations revealed strong and early ER stress that led to disruption of proteostasis accompanied by nitrosative stress and impairment of ERAD[76]. Similarly, levels of UPR markers were elevated in dopaminergic neurons that were generated from iPSCs from patients with PD who had α-synuclein and glucocerebrosidase mutations[77]. In ALS, transcriptional analysis of motor neurons derived from iPSCs from patients carrying a *SOD1* mutation demonstrated that ER stress was high in these cells. Remarkably, UPR alterations observed in motor neurons with *C9orf72* repeat expansions were similar to those in neurons with *SOD1* mutations, highlighting the susceptibility of motor neurons to ER stress[78]. Another study also indicated that decreased survival of iPSC-derived motor neurons with *C9orf72* repeat expansions can be attributed to ER stress in combination with impaired calcium homeostasis and abnormal mitochondrial function[79]. One study has also shown that motor neuron cultures generated from iPSCs from patients with ALS-associated *VAPB* mutations develop chronic ER stress, resulting in secondary oxidative stress, mitochondrial dysfunction, synaptic loss and cell death[80].

iPSC modelling has also provided some insight into other PMDs. In HD, analysis of iPSCs derived from patients has revealed ER calcium dyshomeostasis, which could affect protein folding at the ER[81]. ER stress has also been identified as a salient feature in familial AD and retinitis pigmentosa through the use of iPSC modelling[82–84].

Linking ER stress to neurodegeneration

Chronic activation of the UPR has emerged as a conserved feature among various neurodegenerative diseases on the basis of animal models and post-mortem studies of tissue from patients. Selective neuronal populations seem to be specifically vulnerable to ER stress[85],

Box 2 | **ER proteostasis and ageing**

Perturbed neuronal proteostasis is a salient feature of ageing as well as of protein misfolding disorders[11]. A reduction in the buffering capacity of the proteostasis network during ageing might increase the risk of neurodegeneration by increasing the accumulation of abnormal protein aggregates. Protein aggregates might also propagate via a prion-like mechanism, thereby spreading pathology through the brain[180]. Studies in model organisms indicate that the neuronal unfolded protein response (UPR) has a central role in controlling global proteostasis. For example, a study conducted with *Caenorhabditis elegans* suggest that expression of the ER-stress-induced transcription factor XBP1s in neurons activates global responses in peripheral tissues that improve proteostasis and age resiliance[11,188]. Experiments in mouse models have also indicated that XBP1s expression in the hypothalamus promotes cell-nonautonomous UPR activity in the periphery, including the liver, with effects on global energy metabolism[189].

and studies in mice and humans have shown that ER stress and an impaired UPR are directly associated with neurodegeneration[86]. Over the past decade, genetic and pharmacological manipulation of the UPR have been used to understand the causal link between ER stress and neurodegeneration (FIG. 3). These studies have exposed a multifaceted scenario wherein distinct signalling components of the UPR have specific, and sometimes even opposite, effects on the disease pathophysiology depending on the disease type, the neurons affected and the stage of the disease (TABLE 1). In this section, we discuss the outcomes of key studies that have addressed this functional link, focusing on *in vivo* models and their use for defining optimal targets for disease intervention.

PERK signalling

Generation of small molecules and genetic tools that enable manipulation of PERK signalling in a disease-specific manner in the past 5 years has transformed the study of ER stress and neurodegeneration (TABLE 1). The therapeutic value of PERK modulation has been demonstrated by successsful use of an orally administered PERK inhibitor (GSK2606414) in mice[87], and the successful use of several eIF2α phosphatase inhibitors, including salubrinal[88] (which blocks PPP1R15B and PPP1R15A), guanabenz[89] and its derivative sephin-1 (REF. 90) (which inhibit PPP1R15A). Similarly, a small molecule called ISRIB (integrated stress response inhibitor) efficiently blocks the consequences of eIF2α phosphorylation[91,92].

Initial studies indicated that *Perk* haploinsufficiency accelerates experimental ALS in transgenic mice with a *SOD1* mutation[93]. Consistent with this observation, genetic ablation of GADD34 (REF. 94) or the administration of the eIF2α phosphatase inhibitors guanabenz, sephin-1 or salubrinal[90,95–97] — thereby enhancing blockage of protein translation — had positive effects in improving motor neuron survival and motor performance and delaying the death of the animals. Similarly, eIF2α phosphatase inhibitors reduced neurodegeneration in zebrafish and *Caenorhabditis elegans* models of TDP-43 pathology[98], although another mouse study showed that treatment with guanabenz could accelerate progression of experimental ALS[99]. Ablation of ATF4 expression in mice with *SOD1* mutations protects against ALS, possibly by reducing the levels of apoptosis components, including CHOP[100]. These observations suggest that PERK signalling has a dual role in ALS, promoting survival by repressing protein synthesis upon ER stress, but promoting apoptosis if the stress becomes chronic and irreversible.

A pathological role of ATF4 has been reported in AD. Studies in mouse models and cell culture have shown that local expression of ATF4 in axons leads to transmission of neurodegenerative signals through cell-nonautonomous mechanisms[101]. In experimental PD, *Chop* deficiency was protective[102] and eIF2α phosphatase inhibition with salubrinal alleviated symptoms, but neither affected survival of dopaminergic neurons[103].

Some evidence suggests that ER stress contributes to neurodegeneration in diseases that involve mutation of myelin proteins that causes their misfolding and accumulation at the ER. For example, a pathogenic role of PERK has been demonstrated in mouse models of Charcot–Marie–Tooth disease, a group of inherited disorders that affect the peripheral nerves. Several reports have systematically dissected the effect of PERK signalling on Schwann cell survival and motor deficits; these studies have used *Perk*, *Chop* and *Gadd34* knockout mice, and treatment with salubrinal, and have revealed global neuroprotection and improved motor recovery[104–106] (TABLE 1). Treatment with sephin-1 prevented the development of experimental Charcot–Marie–Tooth disease[90]; one study has brought into question the specificity of guanabenz and its derivative sephin-1 for inhibition of the eIF2α phosphatase subunit PPP1R15A[107], but their actions are neuroprotective regardless of their target. In other demyelinating diseases that affect oligodendrocyte function, such as Pelizaeus–Mezbarcher disease, *Chop* deficiency in mice surprisingly exacerbated the pathology[108], but the mechanism of action was not defined.

IRE1α and XBP1

The role of IRE1α signalling on neurodegeneration has been studied extensively in the context of downstream XBP1 function (TABLE 1), and most studies suggest that XBP1s has a neuroprotective role. Activity of the XBP1 transcription factor has been widely associated with adaptive programmes that alleviate ER stress, so gene therapy in which XBP1s (the active form) is delivered has been tested as a strategy to artificially activate the pro-survival UPR. Delivery of recombinant adeno-associated viruses (AAVs) to express XBP1s in selective brain regions provides outstanding neuroprotection in various mouse models of PMDs (reviewed in REF. 109). For example, local injection of viruses to express XBP1s in the substantia nigra prevented degeneration triggered by PD-inducing neurotoxins[110,111]. Similarly, expression of XBP1s reduced aggregation of mutant huntingtin in the striatum[112] in a mouse model of HD.

Genetic ablation of *Xbp1* specifically in the nervous system of mouse models of ALS and HD has produced surprising observations. Despite the expectation that targeting this major pro-survival mediator of the UPR will accelerate disease progression, conditional genetic deletion of *Xbp1* in the CNS protected against experimental ALS[65]. XBP1 deficiency reduced motor neuron loss and aggregation of mutant SOD1, thereby delaying

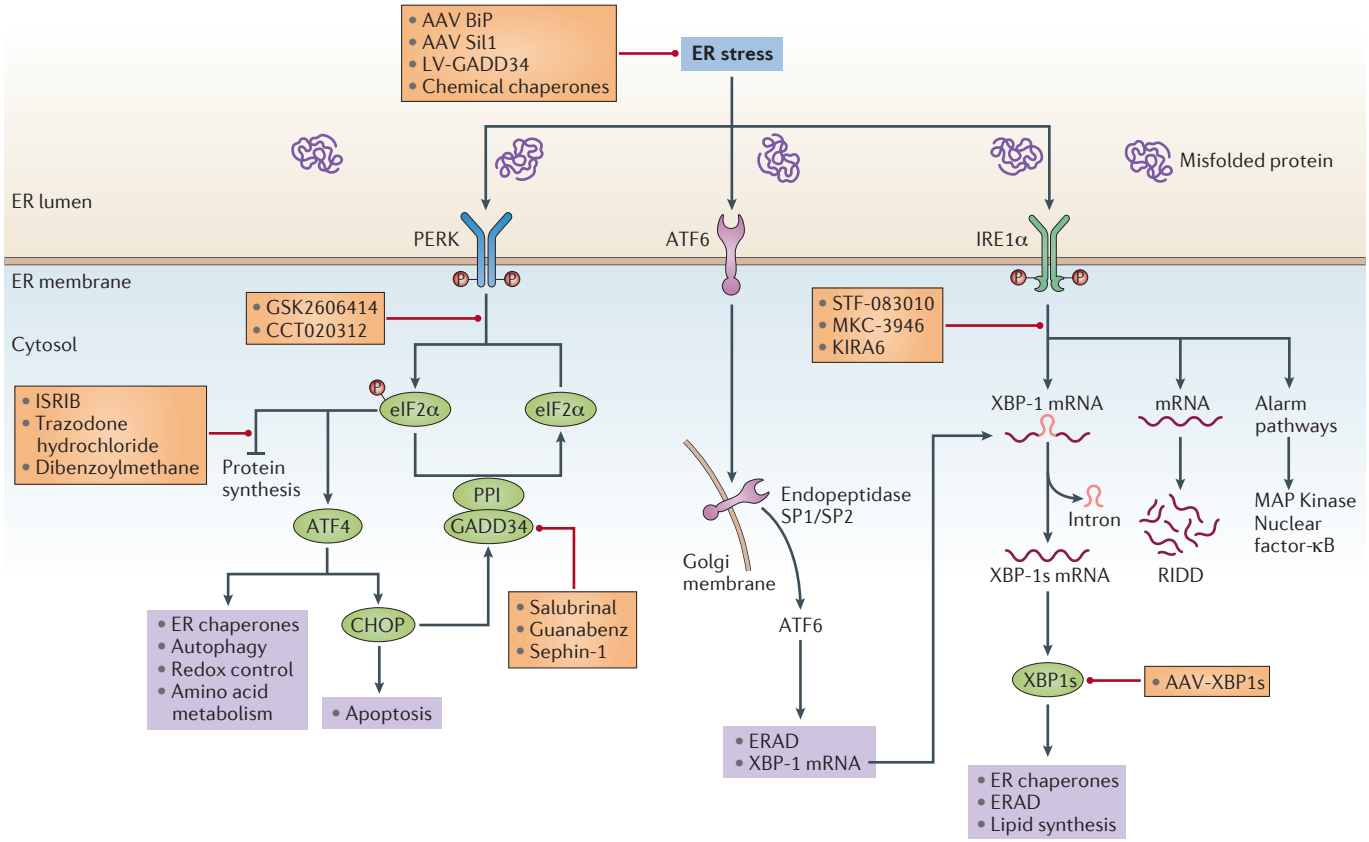

Figure 3 | **Unfolded protein response pathways and interventions.** Accumulation of misfolded proteins in the endoplasmic reticulum (ER) activates the unfolded protein response sensors protein kinase RNA-like ER kinase (PERK), activating transcription factor (ATF) 6 and IRE (inositol-requiring protein) 1α. PERK activation leads to phosphorylation of eIF2α and consequent inhibition of protein translation and expression of the transcription factor ATF4. ATF4 translocates to the nucleus and induces expression of pro-survival genes. ATF4 also controls genes related to apoptosis, including CHOP. CHOP in turn can induce the expression of GADD34, promoting the dephosphorylation of eIF2α under prolonged ER stress. Upon ER stress, ATF6 is transported to the Golgi apparatus, where it is cleaved by endopeptidase S1P and endopeptidase S2P, thereby releasing the cytosolic ATF6 fragment (ATF6f) that operates as a transcription factor. ATF6f induces genes required for ER-associated degradation and modulates XBP1 mRNA levels. Active IRE1α induces splicing of mRNA that encodes XBP1, leading to expression of the active transcription factor XBP1s that upregulates pro-survival processes. IRE1α also associates with the adaptor protein TRAF2 (TNF receptor-associated factor 2) and induces mitogen-activating protein (MAP) kinase activation that modulates autophagy and apoptosis. IRE1α activity also induces regulated IRE1α-dependent mRNA decay (RIDD), which affects various pathways, including lipid biosynthesis, microRNAs, inflammation and apoptosis. Attenuation of ER stress in disease can be achieved by intervention at various points, shown in red using pharmacological or gene therapy approaches. AAV, adeno-associated virus; ERAD, ER-associated degradation; ISRIB, integrated stress response inhibitor ; KIRA6, IRE1α kinase inhibiting RNase attenuator 6; LV, lentivirus; PPI, protein phosphatase 1.

disease onset and death. These beneficial effects were associated with increased autophagy in motor neurons that increased mutant SOD1 clearance[65]. Results of *Xbp1* ablation in a mouse experimental model of HD were almost identical[75], suggesting a tight homeostatic balance between the UPR and autophagy that prevents neurodegeneration. Another report suggested that sustained activation of IRE1α in HD can trigger neuronal loss[113]. By contrast, blocking of XBP1 expression did not modify prion pathogenesis *in vivo*[114].

Studies in fly models also indicate that XBP1s overexpression protects neurons against amyloid-β and tau toxicity[115,116]. In agreement with this concept, targeting of IRE1α expression in the brain in a mouse model of AD indicated a pathological role of the pathway: ablation of IRE1α reduced amyloid-β deposition and fully restored synaptic and cognitive function[59]. These beneficial effects were mapped to the control of APP protein stability by XBP1s, which accelerated the amyloid cascade in the disease model. Studies in *C. elegans* also showed that knocking down XBP1 protects against amyloid-β toxicity, thereby increasing lifespan, via a mechanism that might involve hyperactivation of IRE1α, which controls autophagy through the MAP kinase pathway[117]. These studies are in line with other studies in models of diabetes mellitus and retinal degeneration in which overactivation of IRE1α is pathogenic[118].

In general, XBP1 deficiency in the mouse brain does not result in evidence of ER stress or other conserved molecular alterations to compensate for the absence of XBP1 via the activation of the PERK branch, but the

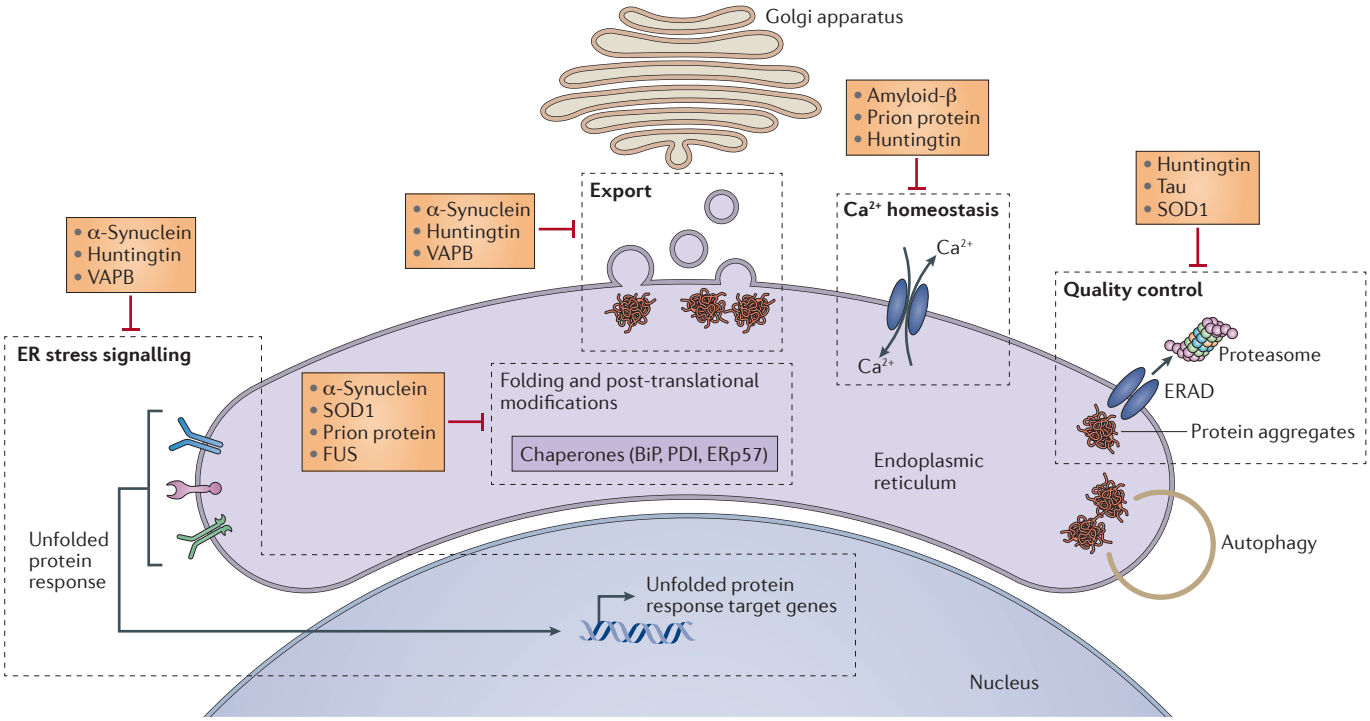

Figure 4 | **Mechanisms that trigger ER stress in neurodegenerative disease.** Correctly folded proteins are processed in the endoplasmic reticulum (ER) and trafficked to the Golgi apparatus for further maturation and distribution to their final destination. Protein folding and maturation at the ER is altered in neurodegenerative disease owing to the effects of protein aggregates on various mechanisms, which include inhibition of protein folding by inhibiting chaperones, interference with the ER-associated degradation pathway, perturbation of ER-to-Golgi trafficking, inhibition of proximal unfolded protein response components, and exacerbation of ER calcium release. ERAD, ER-associated degradation; FUS, fused in sarcoma; PDI, protein disulfide isomerase; SOD1, superoxide dismutase 1; VAPB, vesicle-associated membrane protein-associated protein B.

same is not the true in the substantia nigra. Knockdown of XBP1 in the adult mouse substantia nigra results in chronic ER stress, triggering degeneration of dopaminergic neurons[110]. When XBP1 expression is ablated during brain development, only mild (non-toxic) ER stress occurs in the substantia nigra, but this stress protects from PD via upregulation of the adaptive UPR (upregulation of ER chaperones and autophagy)[110,119]. In agreement with these observations, studies in fly and rat models of PD indicate that treatment with non-toxic doses of tunicamycin, which induces adaptive ER stress signals, also attenuates experimental PD in this way, a mechanism of protection known as ER hormesis[120].

ATF6 signalling

Involvement of ATF6 in neurodegeneration has been poorly studied (TABLE 1). Some studies have been performed in animal models of PD, and have shown that mice with a deficiency of ATF6α are hypersensitive to PD-inducing neurotoxins[121,122]. ATF6α knockout mice exhibit reduced basal levels of BiP in dopaminergic neurons[122], suggesting that activity of the ATF6 pathway is required to maintain proteostasis in this neuronal population. The function of ATF6 is repressed in models of HD, and this repression contributes to disease pathogenesis[123,124]. ATF6α and ATF6β have complementary and redundant activities[125], so the role of this specific branch of the UPR requires further investigation. Given the results of these studies, gain-of-function approaches that involve delivery of ATF6f via gene therapy should be used to define the therapeutic value of targeting this UPR signalling branch for disease treatment.

ER chaperones

UPR target genes whose protein products are chaperones and cofactors involved in protein folding and quality control are also emerging as relevant players in neurodegeneration. One example of these genes is *SIL1*, which encodes an adenine nucleotide exchange factor for BiP. Mutation of *SIL1* causes neurodegeneration associated with abnormal protein aggregation and cerebellar degeneration[126]. *SIL1* mutations have also been linked to Marinesco–Sjögren syndrome, a condition that causes cerebellar ataxia, mental retardation and muscle weakness. Another study showed that the expression level of *SIL1* determines motor neuron susceptibility to ER stress[127]. Gene therapy to increase expression of *SIL1* in the nervous system delayed development of pathology in experimental ALS[127]. Genetic inactivation of BiP itself results in spontaneous degeneration and motor alterations during ageing in association with aggregation of wild-type SOD1 in mice (REF. 128). Interestingly, promoter polymorphisms in the ER chaperones BiP and GRP94 have also been genetically linked

Hormesis
A phenomenon in which an agent that is toxic to a biological system at high doses has beneficial effects on that system at lower doses.

Table 1 | **Functional impact of ER stress in neurodegenerative diseases**

Disease	Model	UPR manipulation	Phenotype	Refs
PERK signalling				
Amyotrophic lateral sclerosis	Mutant *SOD1* Tg mice	*EIF2AK3*$^{+/-}$	Disease exacerbation, enhanced SOD1 aggregation	93
		PPP1R15A$^{+/-}$	Disease exacerbation, enhanced SOD1 aggregation	94
		Salubrinal	Extended life span	97
		Guanabenz	Delayed disease progression, extended life span	95,96
		Guanabenz	Exacerbated disease	99
		Sephin-1	Neuroprotection, delayed disease onset	90
		ATF4 KO	Protection against disease progression	100
Alzheimer disease	PSEN1/APP mice	*EIF2AK3* cKO	Improved learning and memory and LTP	152,153
		siRNA *ATF4*	Neuroprotection	101
	APP mice	ISRIB	No improvement in learning and memory	154
	Tau Tg mice	GSK2606414	Global neuroprotection	145
		CCT020312	Global neuroprotection	150
		Trazodone	Neuroprotection, reduced tau phosphorylation	148
		Dibenzoylmethane	Neuroprotection	148
Charcot–Marie–Tooth disease	Mutant P-myelin mice	Salubrinal	Neuroprotection, Schwann cell survival, motor recovery	106
		CHOP KO	Global neuroprotection	104
		PPP1R15A KO	Global neuroprotection	105
		EIF2AK3 cKO	Global neuroprotection	106
		Sephin-1	Improved motor function and neuroprotection	90
Huntington disease	Mutant Htt Tg mice	*ATF4* KO	No effect on mutant Htt aggregation	75
Parkinson disease	α-synuclein Tg mice	Salubrinal	Neuroprotection	103
	Neurotoxins	*CHOP* KO	Neuroprotection	102
Pelizaeus–Merzbacher disease	Proteolipid mutant mice	*CHOP* KO	Disease exacerbation, oligodendrocyte apoptosis	108
Prion-related disease	Scrapie prion infected mice	Salubrinal	Disease exacerbation	143
		GSK2606414	Reduced neurodegeneration, delayed disease progression	144
		ISRIB	Delayed onset, improved survival	146
		LV-GADD34	Delayed onset	143
		Trazodone	Delayed onset	148
		Dibenzoylmethane	Delayed onset	148
IRE1 signalling				
Amyotrophic lateral sclerosis	Mutant SOD1 Tg mice	*XBP1* cKO	Neuroprotection, reduced SOD1 aggregation	65
Alzheimer disease	APP/PSEN1 mice	*IRE1* cKO	Reduced Aβ load, synaptic function, cognition and glial activation	115,116
		AAV *XBP1s*	Improved neuronal plasticity and behaviour	162
Huntington disease	Mutant Htt Tg mice	*XBP1* cKO	Improved motor performance, reduced Htt	75
		AAV *XBP1s*	Improved motor performance, reduced Htt	112
Parkinson disease	Neurotoxins	AAV *XBP1s*	Reduced dopaminergic neuron loss	110
		AV *XBP1s*	Reduced dopaminergic neuron loss	111
Prion-related disease	Scrapie prion infected mice	*XBP1* cKO	No effect on disease progression or prion replication	114
ATF6 signalling				
Parkinson disease	Neurotoxins	*ATF6* KO	Enhanced neurodegeneration	121,122

AV, adenovirus; AAV, adeno-associated virus; ATF4, activating transcription factor 4; cKO, conditional knockout; ER, endoplasmic reticulum; Htt, huntingtin protein; IRE1, inositol-requiring protein 1; ISRIB, integrated stress response inhibitor; KO, knockout; LV, lentivirus; LTP, long-term potentiation; PERK, protein kinase RNA-like ER kinase; PSEN1, presenilin 1; siRNA, small interfering RNA; SOD1, superoxide dismutase; Tg, transgenic, UPR; unfolded protein response.

to the development of bipolar disorders in humans[129,130]. Overall, these studies suggest that disruption of ER proteostasis can result in severe neuronal dysfunction.

One major folding and quality control pathway for glycoproteins in the ER is mediated by calnexin and calteticulin, lectin chaperones that form a complex with ERp57, which assists with disulfide bond formation. Genetic ablation of calnexin in mice leads to severe demyelination, resulting in reduced axonal conduction velocity and consequent motor defects[131,132]. Similarly, genetic ablation of calreticulin accelerates muscle denervation in ALS[133]. Mutations in the genes encoding PDI and ERp57 have been proposed as risk factors for ALS[134–136]. Conditional deletion of ERp57 in the nervous system triggers motor neuron dysfunction associated with an abnormal structure of neuromuscular junctions and reduced expression of certain synaptic proteins, such as synaptic vesicle protein 2 (REF. 137). Expression of ALS-associated mutant forms of PDI and ERp57 in zebrafish models dramatically alters the morphology of the neuromuscular junction and causes motor deficits[137]. Mutations in other genes of the proteostasis network that are involved in proteasome degradation, such as *UBQLN2*, which encodes ubiquilin 2, and *UCHL1*, which encodes ubiquitin c-terminal hydrolase L1, are also associated with ALS, and their expression in transgenic mice results in ER stress and motor dysfunction[138,139]. All of these studies suggest that motor neurons are highly susceptible to perturbations in ER proteostasis. However, in transgenic mice that overexpressed ERp57 in the nervous system, peripheral nerve degeneration was delayed, although overexpression of ERp57 in a mouse model of PD did not influence dopaminergic neuron survival.[140].

Gene therapy and pharmacological strategies have been developed to improve protein folding and reduce pathological levels of ER stress. Delivery of BiP-overexpressing AAVs to the substantia nigra of rats delayed the progression of PD that is triggered by α-synuclein overexpression, and improved motor performance and dopaminergic neuron survival[141]. The same approach had outstanding effects in a model of retinitis pigmentosa, in which sub-retinal delivery of AAV-BiP diminished photoreceptor apoptosis, attenuated ER stress levels and improved visual function in transgenic rats with mutant rhodopsin[142]. Another therapeutic approach that has been used is administration of chemical chaperones, which are low-molecular-weight compounds that stabilize the protein structure and buffer abnormal protein aggregation. The best characterized chemical chaperones in a disease context are 4-phenyl butyrate (4-PBA), tauroursodeoxycholic acid (TUDCA), and the sugar trehalose, which are all FDA-approved and have good safety profiles in humans. Treatment with various chemical chaperones in mouse models of ALS, HD and PD have indicated that they provide considerable neuroprotection (reviewed in REF. 19), although in most of these studies, ER stress levels were not determined. In summary, the evidence suggests that alterations to the ER folding machinery underlie neuronal dysfunction in various neurodegenerative diseases, indicating that strategies to improve folding could have important therapeutic effects.

Synaptic function and plasticity

Novel evidence suggests that PERK and IRE1α–XBP1 signalling are relevant to neurodegenerative diseases not only because they mediate the attenuation of chronic ER stress, but also because they influence synaptic function through novel mechanisms. In combination, the findings, which are described in the next section, suggest that these pathways alter neuronal function by modulating the expression of important synaptic proteins. Effects of UPR signalling on the establishment of synapses and neuronal plasticity have implications for cognitive and memory function in the context of PMDs.

PERK and synaptic function

Beyond its role as a mediator of ER stress in all cells, evidence suggests that PERK signalling has an additional role in neuronal physiology. Chronic PERK signalling is thought to repress the expression of a cluster of synaptic proteins, leading to altered neuronal plasticity and behaviour, and several studies have indicated that this effect is relevant in neurodegenerative disease.

In prion-infected mice, functional studies have demonstrated that sustained phosphorylation of eIF2α dramatically reduces expression of synaptic proteins[143]. Oral administration of PERK inhibitors to prion-infected animals restored levels of synaptic proteins and delayed the disease course[144]. However, unexpectedly, PERK inhibition did not affect the levels of prion misfolding and aggregation.

In mouse models of tau-mediated FTD, the PERK inhibitor GSK2606414 provided neuroprotection associated with improved synaptic function and neuronal survival that was associated with reduced tau phosphorylation[145]. However, GSK2606414 was associated with adverse effects on pancreatic function owing to a high toxicity of this compound to β cells[146]. Furthermore, the specificity of GSK2606414 has been questioned, as it blocks receptor-interacting serine–threonine protein kinase 1 (RIPK1), a central component of the necroptosis machinery[147]. These adverse effects were not observed when ISRIB was tested to target the pathway in prion-infected animals[146]. A 2017 drug screen identified that two FDA-approved drugs — trazodone hydrochloride and dibenzoylmethane — are de-repressors of translational attenuation mediated by phosphorylated eIF2α[148]. Use of these compounds in mouse models of prion-related disorders and FTD protected against neurodegeneration, and no toxicity was associated with use of clinically-relevant concentrations[148].

In contrast to these studies, another report suggested that PERK signalling is beneficial in FTD. Pharmacological activation of PERK signalling with the drug CCT020312 (REF. 149) improved dendritic spine density in a mouse model of tau-mediated FTD, and this protection was associated with improved cognitive and motor function and attenuated tau pathology. These effects were associated with the phosphorylation of a different PERK substrate, nuclear factor erythroid 2-related factor 2, which is a central component of the antioxidant response[150]. This observation is consistent with the fact that patients with Wolcott–Rallison syndrome,

a rare condition in which diabetes mellitus is caused by loss-of–function mutations in *EIF2AK3*, which encodes PERK, develop early signs of neurodegeneration, including the accumulation of neurofibrillary tangles[151].

In AD, chronic PERK signalling has adverse effects on synaptic function. In mouse models of AD, ablation of PERK improved memory deficits and long-term potentiation[152,153], although inhibition of the integrated stress response with ISRIB (a negative regulator of neuronal plasticity) did not rescue memory deficits in transgenic AD mice[154]. Deletion of PERK in the adult mouse forebrain recapitulates multiple behavioural phenotypes associated with impaired cognition and information processing[155]. However, only in the hippocampus did PERK signalling improve synaptic function by suppressing long-term repression[156]. Overall, many studies indicate that the integrated stress response operates as a global negative regulator of synaptic plasticity through the phosphorylation of eIF2α at basal levels (reviewed in REF. 157).

IRE1α and XBP1 in synaptic function

Studies have suggested that XBP1 has a function in synaptic plasticity and cognition. A polymorphism in the XBP1 promoter reduces total levels of XBP1 (REF. 151) and was initially linked to psychiatric disorders in Japan[158,159], but has more recently been proposed as a risk factor for AD in China[160]. This hypothesis is supported by evidence from a mouse model with a conditional deletion of XBP1, which bypasses the lethality of complete XBP1 deletion[114]. Phenotypic screening of this mouse model identified selective defects in processes related to learning and memory that were associated with altered synaptic transmission in the hippocampus[161].

Studies using XBP1s transgenic mice or local delivery of AAV–XBP1s into the hippocampus, showed that the gain of function improved the basal learning and memory capacity of mice. Gene expression analysis of the hippocampus in XBP1-deficient animals indicated no changes in the expression of typical UPR target genes, but several factors involved in neuronal plasticity were dramatically downregulated[161]. Brain-derived growth factor (BDNF) was among these downregulated factors, and evidence showed that it is directly regulated by XBP1s, suggesting that BDNF mediates this novel activity of the pathway on synaptic plasticity. These observations indicate that XBP1 is part of a molecular network that influences normal synaptic plasticity and memory functions. This concept has been applied to a model of AD, in which expression of XBP1s reversed memory deficits and increased dendritic spine density and synaptic transmission in the hippocampus[162]. Furthermore, evidence in cell culture systems suggests that XBP1 mRNA splicing operates downstream of BDNF signalling[163], regulating the expression of neuropeptides and synaptic genes[164].

Neuroinflammation and axonal damage

All PMDs are characterized by increased neuroinflammation associated with the activation of astrocytes and microglia, and with the infiltration of immune cells into the brain[3,4]. In addition, axonal degeneration is an early pathological event in most PMDs, and occurs before neuronal loss is evident. However, emerging evidence indicates similar roles for ER stress and the UPR in neuroinflammatory disease and neurological injury. Studies suggest that a similar mechanism of neurodegeneration occurs in multiple sclerosis, and that the UPR contributes to the prevention of axonal damage in models of injury and to preservation of oligodendrocyte function under conditions of chronic ER stress in the brain (Supplementary information S1 (table)).

Several studies indicate that inflammatory damage to the nervous system triggers neurodegeneration as a result of chronic ER stress. In experimental autoimmune encephalitis, genetic manipulation of PERK expression or use of eIF2α phosphatase inhibitors has demonstrated that activity of this pathway improves oligodendrocyte function and survival[165–168].

Mechanical damage to the CNS has also been associated with rapid induction of abnormal ER stress levels, followed by motor dysfunction[169]. In the context of experimental spinal cord injury in mice, XBP1 or ATF4 deficiency aggravated motor dysfunction[170]. Remarkably, local delivery of AAVs that encode XBP1s into the injured area attenuated motor deficits and was associated with improved oligodendrocyte survival[170]. Similarly, expression of ATF4 had positive effects on motor recovery after spinal cord injury, and this effect was associated with reduced oligodendrocyte loss[170]. In contusion models of spinal injury in mice, genetic ablation of CHOP or treatment with salubrinal improved oligodendrocyte survival and hindlimb locomotion[171,172]. However, no protection was observed when the stress-inducible eIF2α phosphatase was inhibited with guanabenz or via the deletion of GADD34 (REF. 173). In the context of optic nerve crush, PERK deficiency or treatment of mice with ISRIB reduced axonal degeneration[174]. Finally, ATF6α deficiency modulated ER stress levels after spinal cord injury in mice, but had no effect on motor recovery[175]. Consistent with these observations, administration of chemical chaperones attenuates ER stress levels in animal models of brain damage and spinal cord injury[176,177].

Studies of the PNS have indicated that the UPR improves axonal regeneration. In a model of sciatic nerve crush, XBP1s expression accelerated axonal regeneration[178], and the effect was associated with remyelination and removal of axonal debris, possibly as a result of increased macrophage infiltration and augmented levels of the MCP-1 chemokine. In agreement with this concept, improvement of ER proteostasis by the overexpression of ERp57 delayed peripheral nerve degeneration in mice[140]. Unexpectedly, ATF4 deficiency did not influence axonal degeneration and regeneration after sciatic nerve crush[178].

Together, these studies of inflammatory disease and neuronal injury suggest that chronic ER stress represents a general mechanism of neurodegeneration that is triggered not only by the accumulation of disease-related misfolded proteins but also by the proinflammatory environment observed in neurodegenerative disease. Oligodendrocytes are the main cell type that undergoes ER stress-related damage as a result of these

inflammatory conditions and mechanical injury that is linked to pathological conditions, consistent with the fact that this cell type has high secretory activity and produces large amounts of lipids and myelin proteins.

Perspective and conclusions

The first functional studies to link ER stress with brain damage were published over 18 years ago[29,179]. For almost a decade, ER stress was viewed as a pro-degenerative event that occurred downstream of the primary aetiology, but the availability of genetically modified mice and small molecules to selectively target specific UPR mediators have revolutionized the field, revealing a complex scenario in which the UPR can have distinct outcomes in different diseases and when different signalling components are manipulated. Moreover, the UPR can have opposite effects depending on the disease stage, first operating as a pro-survival factor to sustain proteostasis, but shifting towards the promotion of cell damage during late symptomatic phases. Importantly, ER stress is not only a consequence of deleterious effects on the secretory pathway, but also can trigger a vicious cycle by increasing aggregation of disease-associated proteins. In this context, activation of the UPR can counterbalance protein aggregation by improving protein folding and promoting clearance pathways, such as ERAD and autophagy. As most of the proteins linked with PMDs spread through the brain by a prion-like mechanism[180], the possible role of ER stress in the process of disease propagation through protein misfolding remains to be determined.

Studies indicate that alterations in the function of the ER contribute to the aetiology of several neurodegenerative diseases. The discovery of mutations in genes that encode ER chaperones in ALS and components of the UPR in AD indicates that the mechanisms for surveying and sustaining ER proteostasis are fundamental to the maintenance of function in specific neuronal populations. Remarkably, mutations in *ATF6* were discovered as a cause of achromatopsia, an autosomal recessive disorder associated with colour blindness, photophobia and severely reduced visual acuity; this observation indicates that mutations in central components of the UPR can initiate disease[181–183]. This role becomes highly relevant in light of the new evidence that the UPR modulates synaptic plasticity and neuronal connectivity, and the possible relevance to autism spectrum disorders (ASDs). Studies have also indicated the involvement of UPR signalling in brain development, with effects on neuronal differentiation, neurogenesis and dendrite outgrowth (reviewed in REF. 184), which might have implications for neurodevelopmental diseases. Of note, genetic studies of ASDs have indicated that synaptic dysfunction is one of the molecular pathways underlying this neurodevelopmental disorder. Polymorphisms in neuroligins (NLGNs) have been associated with an increased susceptibility to ASD, and a point mutation in *NLGN3* leads to partial retention of the mutant protein in the ER, which induces UPR signalling[185,186].

Several therapeutic approaches to reducing ER stress are under development. A variety of small molecules are available to stimulate or inhibit PERK signalling, with outstanding results in various preclinical models of neurodegeneration. Small molecules to inhibit IRE1α have also been identified, but have not yet been tested in brain diseases. Targeting the UPR presents challenges, as the pathway has a role in the physiology of various cell types and organs, such as the liver and pancreas, so serious adverse effects are predicted with long-term administration of drugs that target the UPR. Gene therapy is emerging as an attractive alternative to small molecules that could bypass their pleiotropic effects because it enables selective targeting of specific brain regions. Outstanding results have been reported from the use of AAVs to deliver active XBP1s or BiP in several models of neurodegenerative disease, with no adverse effects reported to date. The field of AAV-based gene therapy has undergone enormous expansion since 2015, and several clinical trials are in development in PD and spinal muscular atrophy; the approaches in these trials involve intracerebral injection of AAVs, and no adverse effects have been reported. Furthermore, the expected FDA approval of the first gene therapy for Leber disease will accelerate development in all regulatory aspects of AAV-based therapies.

The long-term consequences of administering the current UPR-targeting drugs and biologics will need to be determined before the field moves forward to using these agents in the clinic. Particular attention should be given to the consequences of manipulating the UPR for basal motor and cognitive functions of the nervous system. In addition, the UPR has a role in sustaining the growth of glioma tumours[187], so stimulation of the UPR could increase the risk of glioma, and this aspect should be evaluated in long-term studies. Future efforts are needed to systematically define the components of the ER proteostasis network that should be targeted for optimal disease intervention.

1. Soto, C. Unfolding the role of protein misfolding in neurodegenerative diseases. *Nat. Rev. Neurosci.* **4**, 49–60 (2003).
2. Aguzzi, A. & O'Connor, T. Protein aggregation diseases: pathogenicity and therapeutic perspectives. *Nat. Rev. Drug Discov.* **9**, 237–248 (2010).
3. Bertram, L. & Tanzi, R. E. The genetic epidemiology of neurodegenerative disease. *J. Clin. Invest.* **115**, 1449–1457 (2005).
4. Taylor, J. P., Hardy, J. & Fischbeck, K. H. Toxic proteins in neurodegenerative disease. *Science* **296**, 1991–1995 (2002).
5. Kaushik, S. & Cuervo, A. M. Proteostasis and aging. *Nat. Med.* **21**, 1406–1415 (2015).
6. Balch, W. E., Morimoto, R. I., Dillin, A. & Kelly, J. W. Adapting proteostasis for disease intervention. *Science* **319**, 916–919 (2008).
7. Hetz, C. & Mollereau, B. Disturbance of endoplasmic reticulum proteostasis in neurodegenerative diseases. *Nat. Rev. Neurosci.* **15**, 233–249 (2014).
8. Frake, R. A., Ricketts, T., Menzies, F. M. & Rubinsztein, D. C. Autophagy and neurodegeneration. *J. Clin. Invest.* **125**, 65–74 (2015).
9. Wang, M. & Kaufman, R. J. Protein misfolding in the endoplasmic reticulum as a conduit to human disease. *Nature* **529**, 326–335 (2016).
10. Oakes, S. A. & Papa, F. R. The role of endoplasmic reticulum stress in human pathology. *Annu. Rev. Pathol.* **10**, 173–194 (2015).
11. Martinez, G., Duran-Aniotz, C., Cabral-Miranda, F., Vivar, J. P. & Hetz, C. Endoplasmic reticulum proteostasis impairment in aging. *Aging Cell* http://dx/doi.org/10.1111/acel.12599 (2017).
12. Cornejo, V. H., Pihan, P., Vidal, R. L. & Hetz, C. Role of the unfolded protein response in organ physiology: lessons from mouse models. *IUBMB Life* **65**, 962–975 (2013).
13. Chow, C. Y., Wang, X., Riccardi, D., Wolfner, M. F. & Clark, A. G. The genetic architecture of the genome-wide transcriptional response to ER stress in the mouse. *PLoS Genet.* **11**, e1004924 (2015).
14. Hetz, C., Chevet, E. & Oakes, S. A. Proteostasis control by the unfolded protein response. *Nat. Cell Biol.* **17**, 829–838 (2015).
15. Dombroski, B. A. *et al.* Gene expression and genetic variation in response to endoplasmic reticulum stress in human cells. *Am. J. Hum. Genet.* **86**, 719–729 (2010).
16. Walter, P. & Ron, D. The unfolded protein response: from stress pathway to homeostatic regulation. *Science* **334**, 1081–1086 (2011).

17. Maurel, M., Chevet, E., Tavernier, J. & Gerlo, S. Getting RIDD of RNA: IRE1 in cell fate regulation. *Trends Biochem. Sci.* **39**, 245–254 (2014).
18. Maly, D. J. & Papa, F. R. Druggable sensors of the unfolded protein response. *Nat. Chem. Biol.* **10**, 892–901 (2014).
19. Hetz, C., Chevet, E. & Harding, H. P. Targeting the unfolded protein response in disease. *Nat. Rev. Drug Discov.* **12**, 703–719 (2013).
20. Pakos-Zebrucka, K. *et al.* The integrated stress response. *EMBO Rep.* **17**, 1374–1395 (2016).
21. Chafekar, S. M. *et al.* Increased Aβ1-42 production sensitizes neuroblastoma cells for ER stress toxicity. *Curr. Alzheimer Res.* **5**, 469–474 (2008).
22. Cornejo, V. H. & Hetz, C. The unfolded protein response in Alzheimer's disease. *Semin. Immunopathol.* **35**, 277–292 (2013).
23. Gouras, G. K. *et al.* Intraneuronal Aβ42 accumulation in human brain. *Am. J. Pathol.* **156**, 15–20 (2000).
24. Fernandez-Vizarra, P. *et al.* Intra- and extracellular Aβ and PHF in clinically evaluated cases of Alzheimer's disease. *Histol. Histopathol.* **19**, 823–844 (2004).
25. Hetz, C., Russelakis-Carneiro, M., Maundrell, K., Castilla, J. & Soto, C. Caspase-12 and endoplasmic reticulum stress mediate neurotoxicity of pathological prion protein. *EMBO J.* **22**, 5435–5445 (2003).
The first paper to describe ER stress in tissue from a patient with neurodegeneration.
26. Torres, M. *et al.* Prion protein misfolding affects calcium homeostasis and sensitizes cells to endoplasmic reticulum stress. *PLoS ONE* **5**, e15658 (2010).
27. Ferreiro, E. *et al.* Involvement of mitochondria in endoplasmic reticulum stress-induced apoptotic cell death pathway triggered by the prion peptide PrP(106–126). *J. Neurochem.* **104**, 766–776 (2008).
28. Coe, H. & Michalak, M. Calcium binding chaperones of the endoplasmic reticulum. *Gen. Physiol. Biophys.* **28**, F96–F103 (2009).
29. Katayama, T. *et al.* Presenilin-1 mutations downregulate the signalling pathway of the unfolded-protein response. *Nat. Cell Biol.* **1**, 479–485 (1999).
30. Terro, F. *et al.* Neurons overexpressing mutant presenilin-1 are more sensitive to apoptosis induced by endoplasmic reticulum-Golgi stress. *J. Neurosci. Res.* **69**, 530–539 (2002).
31. Piccini, A. *et al.* Fibroblasts from FAD-linked presenilin 1 mutations display a normal unfolded protein response but overproduce Aβ42 in response to tunicamycin. *Neurobiol. Dis.* **15**, 380–386 (2004).
32. Steiner, H., Winkler, E., Shearman, M. S., Prywes, R. & Haass, C. Endoproteolysis of the ER stress transducer ATF6 in the presence of functionally inactive presenilins. *Neurobiol. Dis.* **8**, 717–722 (2001).
33. Meier, S. *et al.* Identification of novel Tau interactions with endoplasmic reticulum proteins in Alzheimer's disease brain. *J. Alzheimers Dis.* **48**, 687–702 (2015).
34. Abisambra, J. F. *et al.* Tau accumulation activates the unfolded protein response by impairing endoplasmic reticulum-associated degradation. *J. Neurosci.* **33**, 9498–9507 (2013).
35. Bellucci, A. *et al.* Induction of the unfolded protein response by α-synuclein in experimental models of Parkinson's disease. *J. Neurochem.* **116**, 588–605 (2011).
36. Cooper, A. A. *et al.* α-synuclein blocks ER-Golgi traffic and Rab1 rescues neuron loss in Parkinson's models. *Science* **313**, 324–328 (2006).
Study showing that the earliest deficit that results from α-synuclein expression is a block of trafficking from the ER to the Golgi apparatus, explaining the occurrence of ER stress in PD.
37. Credle, J. J. *et al.* α-Synuclein-mediated inhibition of ATF6 processing into COPII vesicles disrupts UPR signaling in Parkinson's disease. *Neurobiol. Dis.* **76**, 112–125 (2015).
38. Takahashi, R., Imai, Y., Hattori, N. & Mizuno, Y. Parkin and endoplasmic reticulum stress. *Ann. NY Acad. Sci.* **991**, 101–106 (2003).
39. Imai, Y. *et al.* An unfolded putative transmembrane polypeptide, which can lead to endoplasmic reticulum stress, is a substrate of Parkin. *Cell* **105**, 891–902 (2001).
40. Ugolino, J., Fang, S., Kubisch, C. & Monteiro, M. J. Mutant Atp13a2 proteins involved in parkinsonism are degraded by ER-associated degradation and sensitize cells to ER-stress induced cell death. *Hum. Mol. Genet.* **20**, 3565–3577 (2011).
41. Kikuchi, H. *et al.* Spinal cord endoplasmic reticulum stress associated with a microsomal accumulation of mutant superoxide dismutase-1 in an ALS model. *Proc. Natl Acad. Sci. USA* **103**, 6025–6030 (2006).
42. Nishitoh, H. *et al.* ALS-linked mutant SOD1 induces ER stress- and ASK1-dependent motor neuron death by targeting Derlin-1. *Genes Dev.* **22**, 1451–1464 (2008).
The first study to show that mutant SOD1 triggers ER stress by interacting with and blocking activity of the ERAD component Derlin-1 in ALS.
43. Urushitani, M., Ezzi, S. A., Matsuo, A., Tooyama, I. & Julien, J. P. The endoplasmic reticulum-Golgi pathway is a target for translocation and aggregation of mutant superoxide dismutase linked to ALS. *Faseb J.* **22**, 2476–2487 (2008).
44. Atkin, J. D. *et al.* Induction of the unfolded protein response in familial amyotrophic lateral sclerosis and association of protein-disulfide isomerase with superoxide dismutase 1. *J. Biol. Chem.* **281**, 30152–30165 (2006).
45. Farg, M. A. *et al.* Ataxin-2 interacts with FUS and intermediate-length polyglutamine expansions enhance FUS-related pathology in amyotrophic lateral sclerosis. *Hum. Mol. Genet.* **22**, 717–728 (2013).
46. Farg, M. A. *et al.* Mutant FUS induces endoplasmic reticulum stress in amyotrophic lateral sclerosis and interacts with protein disulfide-isomerase. *Neurobiol. Aging* **33**, 2855–2868 (2012).
47. Walker, A. K. *et al.* Protein disulphide isomerase protects against protein aggregation and is S-nitrosylated in amyotrophic lateral sclerosis. *Brain* **133**, 105–116 (2010).
48. Uehara, T. *et al.* S-Nitrosylated protein-disulphide isomerase links protein misfolding to neurodegeneration. *Nature* **441**, 513–517 (2006).
49. Gkogkas, C. *et al.* VAPB interacts with and modulates the activity of ATF6. *Hum. Mol. Genet.* **17**, 1517–1526 (2008).
50. Zhang, Y. J. *et al.* Aggregation-prone c9FTD/ALS poly(GA) RAN-translated proteins cause neurotoxicity by inducing ER stress. *Acta Neuropathol.* **128**, 505–524 (2014).
51. Duennwald, M. L. & Lindquist, S. Impaired ERAD and ER stress are early and specific events in polyglutamine toxicity. *Genes Dev.* **22**, 3308–3319 (2008).
52. Yang, H. *et al.* Huntingtin interacts with the cue domain of gp78 and inhibits gp78 binding to ubiquitin and p97/VCP. *PLoS ONE* **5**, e8905 (2010).
53. El-Daher, M. T. *et al.* Huntingtin proteolysis releases non-polyQ fragments that cause toxicity through dynamin 1 dysregulation. *EMBO J.* **34**, 2255–2271 (2015).
54. Freeman, O. J. & Mallucci, G. R. The UPR and synaptic dysfunction in neurodegeneration. *Brain Res.* **1648**, 530–537 (2016).
55. Hoozemans, J. J. *et al.* The unfolded protein response is activated in pretangle neurons in Alzheimer's disease hippocampus. *Am. J. Pathol.* **174**, 1241–1251 (2009).
56. Hoozemans, J. J. *et al.* The unfolded protein response is activated in Alzheimer's disease. *Acta Neuropathol.* **110**, 165–172 (2005).
57. Stutzbach, L. D. *et al.* The unfolded protein response is activated in disease-affected brain regions in progressive supranuclear palsy and Alzheimer's disease. *Acta Neuropathol. Commun.* **1**, 31 (2013).
58. Unterberger, U. *et al.* Endoplasmic reticulum stress features are prominent in Alzheimer disease but not in prion diseases *in vivo*. *J. Neuropathol. Exp. Neurol.* **65**, 348–357 (2006).
59. Duran-Aniotz, C.*et al.* IRE1 signaling exacerbates Alzheimer's disease pathogenesis. *Acta Neuropathol.* http://dx.doi.org/10.1007/s00401-017-1694-x (2017).
The first report to show that IRE1α signaling is pathogenic in Alzheimer disease mouse models.
60. Nijholt, D. A., van Haastert, E. S., Rozemuller, A. J., Scheper, W. & Hoozemans, J. J. The unfolded protein response is associated with early tau pathology in the hippocampus of tauopathies. *J. Pathol.* **226**, 693–702 (2012).
61. Hoozemans, J. J. *et al.* Activation of the unfolded protein response in Parkinson's disease. *Biochem. Biophys. Res. Commun.* **354**, 707–711 (2007).
62. Hoozemans, J. J., van Haastert, E. S., Nijholt, D. A., Rozemuller, A. J. & Scheper, W. Activation of the unfolded protein response is an early event in Alzheimer's and Parkinson's disease. *Neurodegener Dis.* **10**, 212–215 (2012).
63. Makioka, K. *et al.* Involvement of endoplasmic reticulum stress defined by activated unfolded protein response in multiple system atrophy. *J. Neurol. Sci.* **297**, 60–65 (2010).
64. Atkin, J. D. *et al.* Endoplasmic reticulum stress and induction of the unfolded protein response in human sporadic amyotrophic lateral sclerosis. *Neurobiol. Dis.* **30**, 400–407 (2008).
65. Hetz, C. *et al.* XBP-1 deficiency in the nervous system protects against amyotrophic lateral sclerosis by increasing autophagy. *Genes Dev.* **23**, 2294–2306 (2009).
This study showed that inhibition of XBP1 signalling in ALS ameliorates disease pathology through cross-talk with ER stress and autophagy *in vivo*.
66. Ito, Y. *et al.* Involvement of CHOP, an ER-stress apoptotic mediator, in both human sporadic ALS and ALS model mice. *Neurobiol. Dis.* **36**, 470–476 (2009).
67. Sasaki, S. Endoplasmic reticulum stress in motor neurons of the spinal cord in sporadic amyotrophic lateral sclerosis. *J. Neuropathol. Exp. Neurol.* **69**, 346–355 (2010).
68. Ilieva, E. V. *et al.* Oxidative and endoplasmic reticulum stress interplay in sporadic amyotrophic lateral sclerosis. *Brain* **130**, 3111–3123 (2007).
69. Nardo, G. *et al.* Amyotrophic lateral sclerosis multiprotein biomarkers in peripheral blood mononuclear cells. *PLoS ONE* **6**, e25545 (2011).
70. Prudencio, M. *et al.* Distinct brain transcriptome profiles in C9orf72-associated and sporadic ALS. *Nat. Neurosci.* **18**, 1175–1182 (2015).
71. Yoo, B. C. *et al.* Overexpressed protein disulfide isomerase in brains of patients with sporadic Creutzfeldt-Jakob disease. *Neurosci. Lett.* **334**, 196–200 (2002).
72. Wiersma, V. I. *et al.* Activation of the unfolded protein response and granulovacuolar degeneration are not common features of human prion pathology. *Acta Neuropathol. Commun.* **4**, 113 (2016).
73. Carnemolla, A. *et al.* Rrs1 is involved in endoplasmic reticulum stress response in Huntington disease. *J. Biol. Chem.* **284**, 18167–18173 (2009).
74. Kalathur, R. K. *et al.* The unfolded protein response and its potential role in Huntington's disease elucidated by a systems biology approach. *F1000Res* **4**, 103 (2015).
75. Vidal, R. L. *et al.* Targeting the UPR transcription factor XBP1 protects against Huntington's disease through the regulation of FoxO1 and autophagy. *Hum. Mol. Genet.* **21**, 2245–2262 (2012).
76. Chung, C. Y. *et al.* Identification and rescue of α-synuclein toxicity in Parkinson patient-derived neurons. *Science* **342**, 983–987 (2013).
This study showed that cortical neurons derived from iPSCs from patients with Parkinson disease exhibit nitrosative stress and accumulation of ERAD substrates, which trigger ER stress.
77. Fernandes, H. J. *et al.* ER stress and autophagic perturbations lead to elevated extracellular α-synuclein in GBA-N370S Parkinson's iPSC-derived dopamine neurons. *Stem Cell Rep.* **6**, 342–356 (2016).
78. Kiskinis, E. *et al.* Pathways disrupted in human ALS motor neurons identified through genetic correction of mutant SOD1. *Cell Stem Cell* **14**, 781–795 (2014).
The first study to show that ER stress is a common pathological response in motor neurons derived from rodents and patients with ALS.
79. Dafinca, R. *et al.* C9orf72 hexanucleotide expansions are associated with altered endoplasmic reticulum calcium homeostasis and stress granule formation in induced pluripotent stem cell-derived neurons from patients with amyotrophic lateral sclerosis and frontotemporal dementia. *Stem Cells* **34**, 2063–2078 (2016).
80. Hall, C. E. *et al.* Progressive motor neuron pathology and the role of astrocytes in a human stem cell model of VCP-related ALS. *Cell Rep.* **19**, 1739–1749 (2017).
81. Nekrasov, E. D. *et al.* Manifestation of Huntington's disease pathology in human induced pluripotent stem cell-derived neurons. *Mol. Neurodegener.* **11**, 27 (2016).
82. Kondo, T. *et al.* Modeling Alzheimer's disease with iPSCs reveals stress phenotypes associated with intracellular Aβ and differential drug responsiveness. *Cell Stem Cell* **12**, 487–496 (2013).
83. Jin, Z. B., Okamoto, S., Xiang, P. & Takahashi, M. Integration-free induced pluripotent stem cells derived from retinitis pigmentosa patient for disease modeling. *Stem Cells Transl. Med.* **1**, 503–509 (2012).

84. Tucker, B. A. *et al.* Patient-specific iPSC-derived photoreceptor precursor cells as a means to investigate retinitis pigmentosa.*eLife* **2**, e00824 (2013).
85. Saxena, S. & Caroni, P. Selective neuronal vulnerability in neurodegenerative diseases: from stressor thresholds to degeneration. *Neuron* **71**, 35–48 (2011).
86. Scheper, W. & Hoozemans, J. J. The unfolded protein response in neurodegenerative diseases: a neuropathological perspective. *Acta Neuropathol.* **130**, 315–331 (2015).
87. Axten, J. M. *et al.* Discovery of 7-methyl-5-(1-{[3-(trifluoromethyl)phenyl]acetyl}-2,3-dihydro-1H-indol-5-yl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (GSK2656157), a potent and selective first-in-class inhibitor of protein kinase R (PKR)-like endoplasmic reticulum kinase (PERK). *J. Med. Chem.* **23**, 7193–7207 (2013).
88. Boyce, M. *et al.* A selective inhibitor of eIF2α dephosphorylation protects cells from ER stress. *Science* **307**, 935–939 (2005).
89. Tsaytler, P., Harding, H. P., Ron, D. & Bertolotti, A. Selective inhibition of a regulatory subunit of protein phosphatase 1 restores proteostasis. *Science* **332**, 91–94 (2011).
This report identified guanabenz as a potential drug to target the integrated stress response for therapy in protein misfolding disorders.
90. Das, I. *et al.* Preventing proteostasis diseases by selective inhibition of a phosphatase regulatory subunit. *Science* **348**, 239–242 (2015).
91. Sidrauski, C. *et al.* Pharmacological brake-release of mRNA translation enhances cognitive memory. *eLife* **2**, e00498 (2013).
This study identified the small molecule integrated stress response inhibitor (ISRIB), its targeting of the integrated stress response, and its role in memory consolidation.
92. Sidrauski, C. *et al.* Pharmacological dimerization and activation of the exchange factor eIF2B antagonizes the integrated stress response. *eLife* **4**, e07314 (2015).
93. Wang, L., Popko, B. & Roos, R. P. The unfolded protein response in familial amyotrophic lateral sclerosis. *Hum. Mol. Genet.* **20**, 1008–1015 (2011).
94. Wang, L., Popko, B. & Roos, R. P. An enhanced integrated stress response ameliorates mutant SOD1-induced ALS. *Hum. Mol. Genet.* **23**, 2629–2638 (2014).
95. Wang, L., Popko, B., Tixier, E. & Roos, R. P. Guanabenz, which enhances the unfolded protein response, ameliorates mutant SOD1-induced amyotrophic lateral sclerosis. *Neurobiol. Dis.* **71**, 317–324 (2014).
96. Jiang, H. Q. *et al.* Guanabenz delays the onset of disease symptoms, extends lifespan, improves motor performance and attenuates motor neuron loss in the SOD1 G93A mouse model of amyotrophic lateral sclerosis. *Neuroscience* **277**, 132–138 (2014).
97. Saxena, S., Cabuy, E. & Caroni, P. A role for motoneuron subtype-selective ER stress in disease manifestations of FALS mice. *Nat. Neurosci.* **12**, 627–636 (2009).
This study showed that ER stress is the earliest defect in models of ALS, and that this defect determines the selective vulnerability of motor neurons in ALS.
98. Vaccaro, A. *et al.* Pharmacological reduction of ER stress protects against TDP-43 neuronal toxicity *in vivo*. *Neurobiol. Dis.* **55**, 64–75 (2013).
99. Vieira, F. G. *et al.* Guanabenz treatment accelerates disease in a mutant SOD1 mouse model of ALS. *PLoS ONE* **10**, e0135570 (2015).
100. Matus, S., Lopez, E., Valenzuela, V., Nassif, M. & Hetz, C. Functional contribution of the transcription factor ATF4 to the pathogenesis of amyotrophic lateral sclerosis. *PLoS ONE* **8**, e66672 (2013).
101. Baleriola, J. *et al.* Axonally synthesized ATF4 transmits a neurodegenerative signal across brain regions. *Cell* **158**, 1159–1172 (2014).
102. Silva, R. M. *et al.* CHOP/GADD153 is a mediator of apoptotic death in substantia nigra dopamine neurons in an *in vivo* neurotoxin model of parkinsonism. *J. Neurochem.* **95**, 974–986 (2005).
103. Colla, E. *et al.* Endoplasmic reticulum stress is important for the manifestations of α-synucleinopathy *in vivo*. *J. Neurosci.* **32**, 3306–3320 (2012).
104. Pennuto, M. *et al.* Ablation of the UPR-mediator CHOP restores motor function and reduces demyelination in Charcot-Marie-Tooth 1B mice. *Neuron* **57**, 393–405 (2008).
105. D'Antonio, M. *et al.* Resetting translational homeostasis restores myelination in Charcot-Marie-Tooth disease type 1B mice. *J. Exp. Med.* **210**, 821–838 (2013).
106. Sidoli, M. *et al.* Ablation of Perk in Schwann cells improves Myelination in the S63del Charcot-Marie-Tooth 1B mouse. *J. Neurosci.* **36**, 11350–11361 (2016).
107. Crespillo-Casado, A., Chambers, J. E., Fischer, P. M., Marciniak, S. J. & Ron, D. PPP1R15A-mediated dephosphorylation of eIF2α is unaffected by Sephin1 or Guanabenz. *eLife* **6**, e26109 (2017).
108. Southwood, C. M., Garbern, J., Jiang, W. & Gow, A. The unfolded protein response modulates disease severity in Pelizaeus-Merzbacher disease. *Neuron* **36**, 585–596 (2002).
109. Valenzuela, V., Martinez, G., Duran-Aniotz, C. & Hetz, C. Gene therapy to target ER stress in brain diseases. *Brain Res.* **1648**, 561–570 (2016).
110. Valdes, P. *et al.* Control of dopaminergic neuron survival by the unfolded protein response transcription factor XBP1. *Proc. Natl Acad. Sci. USA* **111**, 6804–6809 (2014).
111. Sado, M. *et al.* Protective effect against Parkinson's disease-related insults through the activation of XBP1. *Brain Res.* **1257**, 16–24 (2009).
112. Zuleta, A., Vidal, R. L., Armentano, D., Parsons, G. & Hetz, C. AAV-mediated delivery of the transcription factor XBP1s into the striatum reduces mutant Huntingtin aggregation in a mouse model of Huntington's disease. *Biochem. Biophys. Res. Commun.* **420**, 558–563 (2012).
113. Hyrskyluoto, A. *et al.* Ubiquitin-specific protease-14 reduces cellular aggregates and protects against mutant huntingtin-induced cell degeneration: involvement of the proteasome and ER stress-activated kinase IRE1α. *Hum. Mol. Genet.* **23**, 5928–5939 (2014).
114. Hetz, C. *et al.* Unfolded protein response transcription factor XBP-1 does not influence prion replication or pathogenesis. *Proc. Natl Acad. Sci. USA* **105**, 757–762 (2008).
115. Casas-Tinto, S. *et al.* The ER stress factor XBP1s prevents amyloid-β neurotoxicity. *Hum. Mol. Genet.* **20**, 2144–2160 (2011).
116. Loewen, C. A. & Feany, M. B. The unfolded protein response protects from tau neurotoxicity *in vivo*. *PLoS ONE* **5**, e13084 (2010).
117. Safra, M., Ben-Hamo, S., Kenyon, C. & Henis-Korenblit, S. The ire-1 ER stress-response pathway is required for normal secretory-protein metabolism in *C. elegans*. *J. Cell Sci.* **126**, 4136–4146 (2013).
118. Morita, S. *et al.* Targeting ABL-IRE1α signaling spares ER-stressed pancreatic β cells to reverse autoimmune diabetes. *Cell Metab.* **25**, 1207 (2017).
119. Mollereau, B. *et al.* Adaptive preconditioning in neurological diseases - therapeutic insights from proteostatic perturbations. *Brain Res.* **1648**, 603–616 (2016).
120. Fouillet, A. *et al.* ER stress inhibits neuronal death by promoting autophagy. *Autophagy* **8**, 915–926 (2012).
121. Egawa, N. *et al.* The endoplasmic reticulum stress sensor, ATF6α, protects against neurotoxin-induced dopaminergic neuronal death. *J. Biol. Chem.* **286**, 7947–7957 (2011).
122. Hashida, K. *et al.* ATF6α promotes astroglial activation and neuronal survival in a chronic mouse model of Parkinson's disease. *PLoS ONE* **7**, e47950 (2012).
123. Naranjo, J. R. *et al.* Activating transcription factor 6 derepression mediates neuroprotection in Huntington disease. *J. Clin. Invest.* **126**, 627–638 (2016).
124. Fernandez-Fernandez, M. R., Ferrer, I. & Lucas, J. J. Impaired ATF6α processing, decreased Rheb and neuronal cell cycle re-entry in Huntington's disease. *Neurobiol. Dis.* **41**, 23–32 (2011).
125. Yamamoto, K. *et al.* Transcriptional induction of mammalian ER quality control proteins is mediated by single or combined action of ATF6α and XBP1. *Dev. Cell* **13**, 365–376 (2007).
126. Zhao, L., Longo-Guess, C., Harris, B. S., Lee, J. W. & Ackerman, S. L. Protein accumulation and neurodegeneration in the woozy mutant mouse is caused by disruption of SIL1, a cochaperone of BiP. *Nat. Genet.* **37**, 974–979 (2005).
127. Filezac de L'Etang, A. *et al.* Marinesco-Sjogren syndrome protein SIL1 regulates motor neuron subtype-selective ER stress in ALS. *Nat. Neurosci.* **18**, 227–238 (2015).
This report identified the role of ER chaperones, such as SIL1, in determining vulnerability of motor neurons to ER stress in ALS.
128. Jin, H., Mimura, N., Kashio, M., Koseki, H. & Aoe, T. Late-onset of spinal neurodegeneration in knock-in mice expressing a mutant BiP. *PLoS ONE* **9**, e112837 (2014).
129. Kakiuchi, C. *et al.* Functional polymorphisms of HSPA5: possible association with bipolar disorder. *Biochem. Biophys. Res. Commun.* **336**, 1136–1143 (2005).
130. Kakiuchi, C. *et al.* Association analysis of HSP90B1 with bipolar disorder. *J. Hum. Genet.* **52**, 794–803 (2007).
131. Kraus, A. *et al.* Calnexin deficiency leads to dysmyelination. *J. Biol. Chem.* **285**, 18928–18938 (2010).
132. Denzel, A. *et al.* Early postnatal death and motor disorders in mice congenitally deficient in calnexin expression. *Mol. Cell. Biol.* **22**, 7398–7404 (2002).
133. Bernard-Marissal, N., Sunyach, C., Marissal, T., Raoul, C. & Pettmann, B. Calreticulin levels determine onset of early muscle denervation by fast motoneurons of ALS model mice. *Neurobiol. Dis.* **73**, 130–136 (2015).
134. Gonzalez-Perez, P. *et al.* Identification of rare protein disulfide isomerase gene variants in amyotrophic lateral sclerosis patients. *Gene* **566**, 158–165 (2015).
135. Kwok, C. T. *et al.* Association studies indicate that protein disulfide isomerase is a risk factor in amyotrophic lateral sclerosis. *Free Radic. Biol. Med.* **58**, 81–86 (2013).
136. Yang, Q. & Guo, Z. B. Polymorphisms in protein disulfide isomerase are associated with sporadic amyotrophic lateral sclerosis in the Chinese Han population. *Int. J. Neurosci.* **126**, 607–611 (2016).
137. Woehlbier, U. *et al.* ALS-linked protein disulfide isomerase variants cause motor dysfunction. *EMBO J.* **35**, 845–865 (2016).
The first study to identify mutations in ER chaperones in ALS.
138. Jara, J. H. *et al.* Corticospinal motor neurons are susceptible to increased ER stress and display profound degeneration in the absence of UCHL1 function. *Cereb. Cortex* **25**, 4259–4272 (2015).
139. Le, N. T. *et al.* Motor neuron disease, TDP-43 pathology, and memory deficits in mice expressing ALS-FTD-linked UBQLN2 mutations. *Proc. Natl Acad. Sci. USA* **113**, E7580–E7589 (2016).
140. Castillo, V. *et al.* Functional role of the disulfide isomerase ERp57 in axonal regeneration. *PLoS ONE* **10**, e0136620 (2015).
141. Gorbatyuk, M. S. *et al.* Glucose regulated protein 78 diminishes α-synuclein neurotoxicity in a rat model of Parkinson disease. *Mol. Ther.* **20**, 1327–1337 (2012).
142. Gorbatyuk, M. S. *et al.* Restoration of visual function in P23H rhodopsin transgenic rats by gene delivery of BiP/Grp78. *Proc. Natl Acad. Sci. USA* **107**, 5961–5966 (2010).
143. Moreno, J. A. *et al.* Sustained translational repression by eIF2α-P mediates prion neurodegeneration. *Nature* **485**, 507–511 (2012).
This report showed that modulating eIF2α phosphorylation restores translation levels and reduces synaptic deficits in prion-related disease in mice.
144. Moreno, J. A. *et al.* Oral treatment targeting the unfolded protein response prevents neurodegeneration and clinical disease in prion-infected mice. *Sci. Transl. Med.* **5**, 206ra138 (2013).
145. Radford, H., Moreno, J. A., Verity, N., Halliday, M. & Mallucci, G. R. PERK inhibition prevents tau-mediated neurodegeneration in a mouse model of frontotemporal dementia. *Acta Neuropathol.* **130**, 633–642 (2015).
146. Halliday, M. *et al.* Partial restoration of protein synthesis rates by the small molecule ISRIB prevents neurodegeneration without pancreatic toxicity. *Cell Death Dis.* **6**, e1672 (2015).
147. Rojas-Rivera, D. *et al.* When PERK inhibitors turn out to be new potent RIPK1 inhibitors: critical issues on the specificity and use of GSK2606414 and GSK2656157. *Cell Death Differ.* **24**, 1100–1110 (2017).
148. Halliday, M. *et al.* Repurposed drugs targeting eIF2α-P-mediated translational repression prevent neurodegeneration in mice. *Brain* **140**, 1768–1783 (2017).
This study identified and characterized the clinically-relevant drugs trazodone hydrochloride and dibenzoylmethane as potential neuroprotective drug targets that target proteostasis.

149. Stockwell, S. R. *et al.* Mechanism-based screen for G1/S checkpoint activators identifies a selective activator of EIF2AK3/PERK signalling. *PLoS ONE* **7**, e28568 (2012).
150. Bruch, J. *et al.* PERK activation mitigates tau pathology *in vitro* and *in vivo*. *EMBO Mol. Med.* **9**, 371–384 (2017).
151. Bruch, J. *et al.* Early neurodegeneration in the brain of a child without functional PKR-like endoplasmic reticulum kinase. *J. Neuropathol. Exp. Neurol.* **74**, 850–857 (2015).
152. Yang, W. *et al.* Repression of the eIF2α kinase PERK alleviates mGluR-LTD impairments in a mouse model of Alzheimer's disease. *Neurobiol. Aging* **41**, 19–24 (2016).
153. Ma, T. *et al.* Suppression of eIF2α kinases alleviates Alzheimer's disease-related plasticity and memory deficits. *Nat. Neurosci.* **16**, 1299–1305 (2013).
This study reported that abnormal PERK activation underlies AD-linked synaptic dysfunction and memory impairment.
154. Johnson, E. C. & Kang, J. A small molecule targeting protein translation does not rescue spatial learning and memory deficits in the hAPP-J20 mouse model of Alzheimer's disease. *PeerJ* **4**, e2565 (2016).
155. Trinh, M. A. *et al.* Brain-specific disruption of the eIF2α kinase PERK decreases ATF4 expression and impairs behavioral flexibility. *Cell Rep.* **1**, 676–688 (2012).
156. Trinh, M. A. *et al.* The eIF2α kinase PERK limits the expression of hippocampal metabotropic glutamate receptor-dependent long-term depression. *Learn. Mem.* **21**, 298–304 (2014).
157. Buffington, S. A., Huang, W. & Costa-Mattioli, M. Translational control in synaptic plasticity and cognitive dysfunction. *Annu. Rev. Neurosci.* **37**, 17–38 (2014).
158. Kakiuchi, C. *et al.* Impaired feedback regulation of XBP1 as a genetic risk factor for bipolar disorder. *Nat. Genet.* **35**, 171–175 (2003).
159. Cheng, D., Zhang, K., Zhen, G. & Xue, Z. The -116C/G polymorphism in XBP1 gene is associated with psychiatric illness in Asian population: a meta-analysis. *Am. J. Med. Genet. B Neuropsychiatr. Genet.* **165B**, 665–672 (2014).
160. Liu, S. Y. *et al.* Polymorphism -116C/G of human X-box-binding protein 1 promoter is associated with risk of Alzheimer's disease. *CNS Neurosci. Ther.* **19**, 229–234 (2013).
161. Martinez, G. *et al.* Regulation of memory formation by the transcription factor XBP1. *Cell Rep.* **14**, 1382–1394 (2016).
The first paper to describe a novel function of XBP1 in regulating learning and memory.
162. Cisse, M. *et al.* The transcription factor XBP1s restores hippocampal synaptic plasticity and memory by control of the Kalirin-7 pathway in Alzheimer model. *Mol. Psychiatry* http://dx.doi.org/10.1038/mp.2016.152 (2016).
163. Hayashi, A. *et al.* The role of brain-derived neurotrophic factor (BDNF)-induced XBP1 splicing during brain development. *J. Biol. Chem.* **282**, 34525–34534 (2007).
164. Hayashi, A., Kasahara, T., Kametani, M. & Kato, T. Attenuated BDNF-induced upregulation of GABAergic markers in neurons lacking Xbp1. *Biochem. Biophys. Res. Commun.* **376**, 758–763 (2008).
165. Clayton, B. L. & Popko, B. Endoplasmic reticulum stress and the unfolded protein response in disorders of myelinating glia. *Brain Res.* **1648**, 594–602 (2016).
166. Lin, W. *et al.* The integrated stress response prevents demyelination by protecting oligodendrocytes against immune-mediated damage. *J. Clin. Invest.* **117**, 448–456 (2007).
167. Lin, W. *et al.* Oligodendrocyte-specific activation of PERK signaling protects mice against experimental autoimmune encephalomyelitis. *J. Neurosci.* **33**, 5980–5991 (2013).
168. Way, S. W. *et al.* Pharmaceutical integrated stress response enhancement protects oligodendrocytes and provides a potential multiple sclerosis therapeutic. *Nat. Commun.* **6**, 6532 (2015).
169. Li, S., Yang, L., Selzer, M. E. & Hu, Y. Neuronal endoplasmic reticulum stress in axon injury and neurodegeneration. *Ann. Neurol.* **74**, 768–777 (2013).
170. Valenzuela, V. *et al.* Activation of the unfolded protein response enhances motor recovery after spinal cord injury. *Cell Death Dis.* **3**, e272 (2012).
171. Ohri, S. S. *et al.* Attenuating the endoplasmic reticulum stress response improves functional recovery after spinal cord injury. *Glia* **59**, 1489–1502 (2011).
172. Ohri, S. S., Hetman, M. & Whittemore, S. R. Restoring endoplasmic reticulum homeostasis improves functional recovery after spinal cord injury. *Neurobiol. Dis.* **58**, 29–37 (2013).
173. Ohri, S. S., Mullins, A., Hetman, M. & Whittemore, S. R. Inhibition of GADD34, the stress-inducible regulatory subunit of the endoplasmic reticulum stress response, does not enhance functional recovery after spinal cord injury. *PLoS ONE* **9**, e109703 (2014).
174. Larhammar, M. *et al.* Dual leucine zipper kinase-dependent PERK activation contributes to neuronal degeneration following insult. *eLife* **6**, e20725 (2017).
175. Saraswat Ohri, S., Hetman, M. & Whittemore, S. R. ATF6α deletion modulates the ER stress response after spinal cord injury but does not affect locomotor recovery. *J. Neurotrauma* http://dx.doi.org/10.1089/neu.2015.3993 (2016).
176. Qi, X., Hosoi, T., Okuma, Y., Kaneko, M. & Nomura, Y. Sodium 4-phenylbutyrate protects against cerebral ischemic injury. *Mol. Pharmacol.* **66**, 899–908 (2004).
177. Mizukami, T. *et al.* Sodium 4-phenylbutyrate protects against spinal cord ischemia by inhibition of endoplasmic reticulum stress. *J. Vasc. Surg.* **52**, 1580–1586 (2010).
An important study that showed that PERK activation in oligodendrocytes reduces the severity of multiple sclerosis.
178. Onate, M. *et al.* Activation of the unfolded protein response promotes axonal regeneration after peripheral nerve injury. *Sci. Rep.* **6**, 21709 (2016).
179. Mattson, M. P., Guo, Q., Furukawa, K. & Pedersen, W. A. Presenilins, the endoplasmic reticulum, and neuronal apoptosis in Alzheimer's disease. *J. Neurochem.* **70**, 1–14 (1998).
180. Soto, C. Transmissible proteins: expanding the prion heresy. *Cell* **149**, 968–977 (2012).
181. Ansar, M. *et al.* Mutation of ATF6 causes autosomal recessive achromatopsia. *Hum. Genet.* **134**, 941–950 (2015).
182. Kohl, S. *et al.* Mutations in the unfolded protein response regulator ATF6 cause the cone dysfunction disorder achromatopsia. *Nat. Genet.* **47**, 757–765 (2015).
183. Chiang, W. C. *et al.* Achromatopsia mutations target sequential steps of ATF6 activation. *Proc. Natl Acad. Sci. USA* **114**, 400–405 (2016).
184. Godin, J. D., Creppe, C., Laguesse, S. & Nguyen, L. Emerging roles for the unfolded protein response in the developing nervous system. *Trends Neurosci.* **39**, 394–404 (2016).
185. De Jaco, A. *et al.* A mutation linked with autism reveals a common mechanism of endoplasmic reticulum retention for the α, β-hydrolase fold protein family. *J. Biol. Chem.* **281**, 9667–9676 (2006).
186. Ulbrich, L. *et al.* Autism-associated R451C mutation in neuroligin3 leads to activation of the unfolded protein response in a PC12 Tet-On inducible system. *Biochem. J.* **473**, 423–434 (2016).
187. Obacz, J. *et al.* Endoplasmic reticulum proteostasis in glioblastoma—From molecular mechanisms to therapeutic perspectives. *Sci. Signal.* **10** (2017).
188. Taylor, R. C. & Dillin, A. XBP-1 is a cell-nonautonomous regulator of stress resistance and longevity. *Cell* **153**, 1435–1447 (2013).
189. Williams, K. W. *et al.* Xbp1s in Pomc neurons connects ER stress with energy balance and glucose homeostasis. *Cell Metab.* **20**, 471–482 (2014).

Acknowledgements
C.H. is supported by the FONDAP program 15150012, the US Office of Naval Research-Global (ONR-G) N62909-16-1-2003, the Millennium Institute P09-015-F, FONDEF ID16I10223, FONDEF D11E1007, the US Air Force Office of Scientific Research FA9550-16-1-0384, CONICYT-Brazil 441921/2016-7, the ALS Therapy Alliance 2014-F-059, the Muscular Dystrophy Association 382453, the Michael J Fox Foundation for Parkinson's Research – Target Validation grant No 9277, FONDECYT no. 1140549, and the ALSRP Therapeutic Idea Award AL150111. S.S. is supported by the Synapsis Foundation, Stiftung UNISCIENTIA, the Frick foundation for ALS research, Swiss National Science Foundation and European Research Council 725825.

Author contributions
Both authors contributed equally to all aspects of the manuscript,

Competing interests statement
The authors declare no competing interests.

Publisher's note
Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

SUPPLEMENTARY INFORMATION
See online article: S1 (table)
ALL LINKS ARE ACTIVE IN THE ONLINE PDF