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The Stress of Lung Aging: Endoplasmic Reticulum and Senescence Tête-à-Tête

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Beyond the structural changes, features including the dysregulation of endoplasmic reticulum (ER) stress response and increased senescence characterize the lung aging. ER stress response and senescence have been reported to be induced by factors like cigarette smoke. Therefore, deciphering the mechanisms underlying ER and senescent pathways interaction has become a challenge. In this review we highlight the known and unknown regarding ER stress response and senescence and their cross talk in aged lung.

cellular senescence; COPD; endoplasmic reticulum stress; lung aging; unfolded protein response

Introduction

Improvement in healthcare systems and advancement in medical sciences, while immensely beneficial, have contributed considerably to increases in aging populations worldwide. Aging is a biological process characterized by progressive decline of cell/tissue function (1). Concurrently, diseases of aging across multiple organ systems have also seen an increase in incidence, as have diseases that show greater occurrence with aging. In this regard, the lung is no exception, with age-related chronic lung diseases, including chronic obstructive pulmonary disease (COPD), showing biological, structural, and physiological similarities to the aged lung but at an accelerated pace and with greater changes that lead to detrimental function that does not occur with normal lung aging (2–4). Furthermore, there is now increasing recognition that proximal airway diseases such as asthma in the elderly are a distinct entity representing an increasing healthcare burden (5). In these diseases, beyond functional effects such as airway hyperreactivity, remodeling of the airways and parenchyma resulting from altered cell proliferation and extracellular matrix (ECM) formation is one of the key features that is also observed in normal aging lung (6–9) and contributes to impairment of gas exchange and overall lung function decline (10–12). Thus it becomes important to understand common mechanisms in the lung that contribute to normal aging versus aging-associated diseases, toward development of novel biomarkers and therapeutics in a vulnerable population.

A multitude of mechanisms are likely involved in the aging process, with cell/tissue and context-specific differences in their relative contributions. Processes such as genomic instability, epigenetics, mitochondrial structure/function, nutrient sensing, proteostasis, immune

responses, cellular senescence, and stem cell depletion are all considered key contributors (13). Importantly, these processes are also interlinked making it difficult to appreciate their individual and net contributions to the aging process or in disease pathophysiology. Nonetheless, loss of proteostasis and cellular senescence have been identified as key contributors to lung aging (13) and are potentially relevant to lung remodeling in the context of disease. Furthermore, there is increasing information linking proteostasis and senescence in the context of aging. Their appeal further lies in the emerging approaches to targeting dysfunctional proteostasis and senescence toward alleviating their detrimental effects. Senescence is known to be triggered by several hallmarks of aging including DNA damage, shortening of telomeres, and mitochondrial dysfunction (12, 14, 15). However, recent investigations have revealed the possible involvement of loss of proteostasis in senescence activation as well. In turn, oxidative stress and epigenetic alterations and the varied responses to these stresses are the main drivers of loss of proteostasis (16) that further leads to an increased amount of unfolded proteins in the endoplasmic reticulum (ER) lumen that subsequently induces ER stress. In response to ER stress, an unfolded protein response (UPR) is initiated to maintain proteostasis. However, during aging, loss of proteostasis is partly the consequence of an imbalance between increased ER stress and decreased UPR, resulting in a chronic ER stress (17). Beyond the impaired ER stress response in aging, an increased number of senescent cells is observed in aged tissues including lung (12).

This review will provide an overview about current knowledge in regards to the cross talk between ER stress response and senescence in lung aging and clearly introduce the fields that still need to be explored to create a better understanding of age-related lung pathology and pathophysiology, with a particular focus on COPD.

ER Stress and Relation with Age

The ER is responsible for the biosynthesis and trafficking of approximately one-third of the entire proteome of the cell. From their biosynthesis to their trafficking and degradation, proteins are controlled through different mechanisms including molecular chaperone networks and the ubiquitin-proteasome system (UPS) in the ER (3, 13, 18). The functioning of these mechanisms has been shown to be impaired with age in different organs including skin, liver, heart, brain, spleen, and lung (3, 19, 20). In the ER, chaperones such as calnexin, calreticulin, and binding immunoglobulin protein (BiP) [also known as glucose-regulated protein-78 (GRP78)] recognize and bind nascent and misfolded proteins to prevent their aggregation (21, 22). The decline of these chaperones with age may lead to an increased amount of unfolded proteins and protein aggregates. A study investigating the expression of calnexin in human fibroblasts from two different passage numbers (appraising passage 20 and 40; defined as young and old cells, respectively) found that old fibroblasts expressed less calnexin compared with young fibroblasts (23). Additionally, lower expression of BiP in alveolar epithelial type II cells (AEC2) from old mice (17–19 mo) compared with young mice (2 mo) has been reported (24). Furthermore, BiP, protein disulfide isomerase (PDI), and calnexin expression at both mRNA and protein level were decrease with age in rat hippocampus (25). The heat shock protein 70 (hsp70) is a molecular chaperone that supports protein folding by preventing aggregation, promoting folding to native state and catalyzing the solubilization and the refolding of aggregated proteins, which are a cluster of unfolded proteins (26). Lung fibroblasts from old human donors express less hsp70 at mRNA and protein level compared with lung fibroblasts from young donors (27). Along the same lines, the level of hsp70 in lung fibroblasts at late passages is decreased compared with lung fibroblasts from earlier passages (27). These emerging data show aging-related decline in chaperone function in lung cells that may have a downstream impact on the ER and its functionality.

As more than 30% of newly synthesized proteins are misfolded, the protein folding process is considered an inaccurate process (28). However, over 90% of misfolded and damaged proteins are cleared through the UPS, which is a highly selective process as it degrades exclusively ubiquitinated proteins (13). As the protein quality control network of the cell, the UPS is crucial for the degradation of misfolded or unfolded and damaged proteins (19, 29). However, this mechanism becomes impaired with age (13). UPS impairment has been shown in aged heart, spinal cord, and skin (30–32). For example, proteasome activity, in dermal fibroblasts from subjects aged from 20 to 82 yr and T cells from peripheral blood from young

(21–30 yr) and old (65–85 yr) healthy human donors, was downregulated with age in both cases (31, 33). In the rat lung, proteasome activity has been reported to be decreased in 2-yr-old aged male rats compared with 2-wk-old male rats (34). However, another study showed that the proteasome activity is not impaired in the lungs of healthy aged mice (35) but that the caspase-like proteasome activity is significantly reduced in lung tissues from aged mice compared with young mice (35). Additionally, an increase of ubiquitinated proteins was demonstrated in the hippocampus (25) and heart (30) of old rats compared with the young rats. Thapsigargin and tunicamycin are commonly used ER stress triggers in *in vivo* and *in vitro* experiments. These compounds each induce ER stress in a specific manner. Thapsigargin inhibits specifically the sarcoplasmic/endoplasmic reticulum calcium ions ATPase (36, 37), resulting in loss of the activity of calcium-dependent ER chaperones such as calnexin. In contrast, tunicamycin induces ER stress by inhibiting the earlier step of the biosynthesis of glycoproteins in the ER (36, 38), provoking the accumulation of unfolded glycoproteins.

Beyond the impairment of mechanisms ensuring the protein quality control in the ER, genetic mutations, errors in transcription and translation, toxic compounds and dysregulation of cellular redox systems, intracellular calcium efflux, and viral infections result in a considerable increasing amount of misfolded proteins in the ER (39, 40). These events may result in ER stress activation in the aged lung (FIGURE 1). Taken together, apart from a few *in vitro* studies, chaperone networks and UPS alterations with age in the lung remain an underexplored area. Therefore, further studies are needed to describe and understand the impact of the changes occurring with age in UPS and chaperone networks in the human lung.

ER Stress and UPR Signaling in Aging

Following activation of ER stress through different stimuli, the UPR signaling pathway is initiated to restore proteostasis (22, 41, 42). UPR signaling pathways consist of three sensors including inositol-requiring enzyme-1 (IRE1), protein kinase RNA-like ER kinase (PERK), and activating transcription factor-6 (ATF6), which are ER-transmembrane proteins recognized also as ER stress markers. These ER stress markers are maintained in inactivated states through their interaction with BiP, an ER chaperone that contains a NH₂-terminal ATPase and COOH-terminal substrate domain (40). The BiP-ATP complex has an increased binding capacity for low affinity substrates including IRE1, PERK, and ATF6 and maintains them in an inactivated state under nonstress conditions (17). The accumulation of unfolded or misfolded proteins in the ER lumen leads to the dissociation of BiP-ATP from ER stress markers and hydrolyzation of ATP by BiP

FIGURE 1. Potential triggers of endoplasmic reticulum (ER) stress in aged lung

Different factors including impairment of ubiquitin-proteasome system and molecular chaperone system, the elevated level of genetic mutations and weakening of transcription and translation machinery, the increased susceptibility to viral infection due to impairment of defense mechanisms, the imbalance of cellular redox system, and the intracellular calcium efflux are the principal drivers of ER stress in aged lung.

ATPase domain to BiP-ADP complex, which binds to hydrophobic peptides contained in the unfolded proteins (40, 43). Thus the dissociation of BiP-ATP from ER stress markers results in activation of UPR signaling pathways, establishing BiP as the principal regulator of ER stress (40, 43). IRE1 and PERK are activated through autophosphorylation of their cytosolic serine/threonine kinase domains, while ATF6 is first translocated to the Golgi apparatus where it is cleaved in the luminal domain and the phospholipid bilayer by site-1 protease (S1P) and metalloprotease site-2 protease (S2P), respectively (40, 44, 45). PERK activation induces inhibition of protein translation and an antioxidant response, while activation of IRE1 and ATF6 promotes the transcription of genes that increase ER folding capacity and function (13). Additionally, IRE1 activation induces degradation of misfolded proteins through the activation of UPS (17). However, when these first measures fail to reestablish homeostasis in the ER, the UPR-mediated apoptosis pathway is activated through PERK, IRE1, and/or ATF6 signaling pathways (46). The activation of the UPR signaling pathways and their downstream effectors is represented schematically in [FIGURE 2](#).

Activation of UPR signaling pathways leads to the expression of various downstream effectors including BiP, calnexin, calreticulin, activator transcription factor-4 (ATF4), C/EBP homologous protein-10 (CHOP), GRP94, X-box binding protein (XBP) 1 spliced variant, and the PDI family (22). These downstream effectors, which contribute to the reestablishment of proteostasis have been reported to decline with age in several mammalian organs (22). In mice sleep deprivation as a stressor results in higher levels of phosphorylated (p) PERK and eukaryotic translation initiation factor 2a (eIF2a), but lower BiP expression in the brains of young 3-mo-old mice compared with those of 22- to 24-mo-old mice (47). Transcriptomic analysis of ER stress and UPR markers in human skeletal muscles after exercise revealed PERK, BiP, eIF2a, and XBP1 were decreased with age (48), PERK and CHOP were increased, whereas p-eIF2a, ATF4, and BiP were decreased in several organs including brain, heart, spleen, liver, and lung from >18-mo-old rats compared with 1-mo young rats (49). However, changes in ER stress response occurring in different human lung cell types with age remain unstudied. Although the focus

FIGURE 2. The 3 signaling pathways of the endoplasmic reticulum (ER) stress response

1) The increase of unfolded proteins in ER lumen leads to the dissociation of binding immunoglobulin protein (BiP)-ATP from ER stress markers including inositol-requiring enzyme-1 (IRE1), protein kinase RNA-like ER kinase (PERK), and activating transcription factor-6 (ATF6), followed by the dephosphorylation of BiP-ATP to BiP-ADP, which has a higher binding affinity to hydrophobic regions of the unfolded proteins. 2) BiP-ATP dissociation from ER stress markers engenders their activation. IRE1 α , and PERK are activated through autophosphorylation, whereas ATF6 activation is effective after its cleavage in the Golgi apparatus by site-1 and site-2 protease (S1P and S2P). 3) IRE1 α , and PERK signaling inhibit protein synthesis. An UPR-specific transcription (ER chaperones, ERAD machinery, and/or trafficking) is initiated by IRE1 α and ATF6 to boost the folding capacity of ER to restore the homeostasis. When they fail to reestablish the proteostasis upon sustained ER stress, the death pathway is activated through PERK signaling. XBP1, X-box binding protein 1; eIF2 α , eukaryotic translation initiation factor 2 α .

of this review is lung, we have reported the changes in ER stress and ER stress response in other organs to emphasize the role of aging on inducing changes in these events and highlight how these events may be reflected in the aged lung.

Cellular Senescence in Lung Aging

Cellular senescence is involved in normal embryonic development, wound healing and injury repair, and organismal aging (50). Cellular senescence is also

one of the key features of aging (1, 13). Hayflick and Moorhead (51), in 1961, were among the first to describe senescence as proliferation arrest, in vitro, resulting from the shortening of telomeres following a limited number of cell divisions. This discovery highlighted senescence as a consequence of cell aging. Depending on the factors that engender senescence, two types can be distinguished: replicative and acute/premature senescence (50–52). The original descriptions by Hayflick and Moorhead (51) represent

replicative senescence, whereas acute/premature senescence is caused by a myriad of stressors including oxidative stress, DNA damage, epigenetic stress, mitochondria dysfunction, proteotoxicity, and metabolic imbalance (50–54). The accumulation of these stressors combined with a decline in defense/protective mechanisms with age, can thus contribute to an increasing number of senescent cells within aging organs. Senescent cells are characterized by long-lasting arrest of cell proliferation, resistance to apoptosis, and sustainable and increased metabolic activity (52, 55, 56). In this regard, a number of studies have explored markers to unambiguously identify senescent cells, although presently there is no unique marker to do so. Furthermore, the profile of senescent cell markers can differ based on cell type and context, and thus multimodal approaches are needed to identify and characterize them. Key features of senescent cells include upregulated expression of cell cycle inhibitors, e.g., cyclin-dependent kinase inhibitors 2A (p16/CDKN2A) and 1A (p21/CDKN1A), up-regulated senescence-associated β -galactosidase (SA- β -GAL) activity, laminin B1 silencing, and increased frequency of telomere-associated DNA damage foci (TAF) and specific senescence-associated secretory phenotype (SASP) components, which consist of cytokines, chemokines, growth factors, proteases, and ECM proteins (52, 57). Indeed, it is now recognized that the effects of senescent cells are mediated through the SASP that impacts on neighboring naïve cells to induce and promote dysfunction (58).

A recently published study showed the upregulation of senescence biomarkers in aged tissues including blood vessels, intestine, liver, kidney, spleen, pancreas, skin, and lung (59). Senescence biomarkers including p16 and p21 are upregulated in the lungs of aged mice (59). Human mesenchymal stem cells (hMSC) show increased level of SA- β -GAL activity in late passage compared with hMSC in earlier passages (60). Additionally, SASP components such as interleukin (IL) 1 α , IL1 β , IL6, tumor necrosis factor- α (TNF α), and the chemokine (C-X-C motif) ligand 1 (CXCL1) and the inflammation index are significantly upregulated in lung homogenates from old mice compared with young mice (61). Lysates from old mice and human lungs also show an upregulation of p16, chromatin-associated protein γ H2x, and the tumor repressor protein Retinoblastoma (Rb) (61). Furthermore, activation of two senescence-inducing pathways, p53/p21 and p16/pRb, has been observed in the lungs of mid-aged and aged mice (62). The contribution of cellular senescence has been extensively reviewed in lung aging, COPD and idiopathic pulmonary fibrosis (IPF) (58, 63, 64). Given cell type and context differences in senescence, it becomes important to also evaluate senescence in different lung cell types such as fibroblasts, airway smooth muscle cells (ASMCs), and airway epithelial cells, and particularly in humans, as paracrine effects of senescent cells on these different cell types

via SASP are crucial for understanding normal lung aging versus dysfunction in the context of disease. Furthermore, senescent cells via SASP may contribute to even greater numbers of senescent cells in naïve tissues with age. Thus understanding the biology of senescent cells in the lung becomes all the more important.

ER Stress Response and Senescence in COPD

COPD is an age-related lung disease that shares many of the features of normal lung aging. Indeed, structural remodeling of the normal aging lung parenchyma is well-known to represent senile emphysema without parenchymal destruction as occurs in COPD per se. Additionally both COPD and aging lungs show narrowing of airways, loss of proteostasis, and increased senescence (2). The importance of UPR in the pathogenesis of COPD has been shown in different studies (16, 41). Factors that trigger ER stress such as dysregulated chaperone networks and impaired UPS activity in aged lung have been observed in COPD lungs. UPS activity is impaired in severe COPD patients, characterized by accumulation of ubiquitinated proteins, which serve as an indicator for an increased level of unfolded and/or misfolded proteins leading to ER stress induction and UPR activation (65). Furthermore, UPR markers and their downstream effectors including ATF6, IRE1 α , p-PERK, p-eIF2 α , ATF4, spliced variant of X-box binding protein (XBP1s), BiP, glucose-regulated protein 94 (GRP94), ER degradation-enhancing α -mannosidase-like (EDE), and CHOP are all upregulated at both the mRNA and protein levels in human primary epithelial cells isolated from the lungs of COPD patients (66). Furthermore, Aggarwal and colleagues (67) reported a significant increase of BiP level in plasma from COPD IV compared with control and COPD II/III. More interesting, it has been demonstrated that rodent model of COPD exposed to the halogen gas bromide developed emphysema and showed an increased level BiP, p-PERK, ATF4, and CHOP but an inhibition of ER stress with salubrinol, which is a specific inhibitor of eIF2 α dephosphorylation, prevented the development of emphysema (67). As the leading cause of COPD, cigarette smoke (CS) effects on these pathways are obviously relevant. Several studies have shown that cigarette smoke extract (CSE) induces ER stress and UPR in human and mouse epithelial cells (68–70). Additionally, it has been shown that the proteasome activity is dysregulated in the lungs of mice and human epithelial cell lines A549 and 16HBE upon CSE exposure (71). Bronchoalveolar lavage fluid (BALF) samples from healthy smokers showed an increased BiP expression compared with nonsmokers samples, and human airway epithelial cells from biopsies showed an increased BiP level upon cigarette smoke exposure (72). Deficits of the antioxidant enzyme

glutathione peroxidase (GPx-1), which is regulated by the nuclear factor erythroid 2-related factor 2 (NRF2), a downstream effector of PERK-pathway, have been shown in COPD lungs (73), therefore suggesting the contribution of UPR pathways in COPD pathogenesis.

Beyond the ER stress response in COPD lungs and models, senescence has become one of the major focuses in COPD research. As with senescence, COPD pathogenesis is driven, among others, by persistent oxidative stress caused by excessive CS and noxious gases exposure (74). The persistent oxidative stress resulting from an imbalance between oxidant and antioxidant systems that is a major driver of COPD pathogenesis (75) is also seen contributing to senescence through DNA damage and telomere shortening (76). Interestingly, shortening of telomeres in C57BL/6J mice increases susceptibility to development of emphysema (77). Furthermore, circulating leukocytes from COPD patients show shorter telomeres compared with healthy subjects (78, 79). Other features of senescence such as p16/CDKN2A, and p21/CDKN1A and lipofuscin have been found to be significantly increased, and telomere length significantly reduced, in AEC2 and endothelial cells from emphysematous lung tissues of COPD-patients compared with lung tissues from smoker and nonsmoker controls (80). Furthermore, p16/CDKN2A, p21/CDKN1A, SA- β -GAL, and the marker of DNA damage γ -H2A.X are increased in primary parenchymal fibroblasts derived from COPD patients compared with matched non-COPD controls (81). Furthermore, expression of p21/CDKN1A, SA- β -GAL, γ -H2A.X, IL6, and IL8 are significantly increased, while p16/CDKN2A is significantly decreased, in paraquat-induced cellular senescence in COPD and control fibroblasts compared with nontreated COPD and control fibroblasts (81). To summarize, a change in the level of UPR markers, impaired UPS, and increased level of senescent cells were demonstrated in COPD. COPD factors such as CS have been revealed as inducers of ER stress and senescence in COPD, but it remains unclear through which signaling pathways these factors activate senescence. This indicates a possible cross talk between ER and senescent pathways. Deciphering the mechanisms underlying this cross talk will help clarifying the nature of their interaction.

ER Stress Response and Cellular Senescence Cross Talk

Oxidative stress is recognized as a key factor contributing to cellular senescence through several event including loss of proteostasis in aging (82). However, besides oxidative stress other factors described in [FIGURE 1](#) can induce loss of proteostasis with age. The demonstration of increased ER stress with decreased UPR and an increase in cellular senescence in aged

lung triggers the question whether impairment of the ER stress response and increased cellular senescence are linked, and whether one drives the other versus being mutually interactive. Exposure of mouse embryonic fibroblasts (MEFs) to thapsigargin leads to cell cycle arrest at G₂/M phase by inhibiting cyclin B1 expression through PERK signaling pathway (83). Hereby, PERK activation leads to the phosphorylation of eIF2 α , which induces expression of growth arrest and DNA-damage-inducible protein 45 α (GADD45 α) (83), a protein involved in cellular senescence. Inhibition of GADD45 α by GADD45 α siRNA reduces cyclin B1 level indicating the essential role that GADD45 α plays in cyclin B1 expression and in turn cell cycle regulation (83). In this model, an increase of GADD45 α expression following exposure to thapsigargin is observed (83); however, this does not represent a cellular senescence model. However, microinjection of GADD45 α expression vector into primary human fibroblasts results in cell cycle arrest at G₂/M phase (84). Furthermore, it has been demonstrated that GADD45 α is involved in cellular senescence through tumor protein p53, which binds preferably to the promoters of cell growth arrest genes p21 and GADD45 α in senescent human fibroblasts (85). Another member of GADD family, GADD34, has also been reported to be involved in senescence. GADD34-induced p21 expression in wild-type MEFs and GADD34-knockout in MEFs reveal an impairment of p21 expression; however, overexpression of GADD34 in knockout MEFs lead to cell growth arrest and increased p21 level (86). Thapsigargin induces GADD34 expression in MEFs (87), and GADD34 expression is activated via PERK-ATF4 signaling pathways (88).

A recent study has shown that normal human dermal fibroblasts (NHDFs) undergo cellular senescence upon 3 days exposure to ER stressors including thapsigargin, tunicamycin, and dithiothreitol (DTT) (89). Moreover, p53-binding protein 1 (53BP1) foci, which serves as a marker for DNA double-strand break (DSB), as it is involved in DSB repair, is detected in the nucleus of NHDFs exposed to thapsigargin, tunicamycin, or dithiothreitol- but not in untreated NHDFs, indicating DNA damage (89). The silencing of ATF6, IRE1 α , and PERK gene does not influence the growth rate of senescent NHDFs. Interestingly, upon replicative senescence, siRNA-ATF6 α and siRNA-IRE1 α but not siRNA-PERK-treated NHDFs display a significant reduction of SA- β -Gal and IL-6 level compared with control cells (89). Senescent NHDFs with silenced ATF6 regain their original fusiform shape resembling the shape of exponential growing cells (89). In the human breast adenocarcinoma cell line MCF-7, oxidative stress triggers cellular senescence through the UPR arm ATF6 and ATF6 silencing in MCF-7 cells prevents them from undergoing cellular senescence upon oxidative stress-mediated ER stress (90). Additionally, stressed cells display increased p21 level and SA- β -GAL activity whereas

ATF6-knockdown MCF-7 cells do not (90). The UPR arm ATF6 also seems to be involved in UV irradiation-mediated cellular senescence in MCF-7 cells (90). NHDFs exposed for 4 days to DTT show significantly increased levels of SA- β -GAL activity and an enlarged of cell surface compared with controls (91). Additionally, it has been shown that DTT induces senescence through the ATF6-cyclooxygenase 2 (COX2)/PTGS2-prostaglandin E2 (PGE2) axis and therefore regulates cell size and SA- β -GAL activity (91). However, with DTT-mediated ER stress, COX2 and PGE2 expression is controlled by both UPR arms, ATF6 and IRE1 α (91). ATF6 and COX2-knockdown restore normal cell size and reduce SA- β -GAL activity (91), but since IRE1 α also regulates COX2 and PGE2 expression, it will be appealing to examine the role of IRE1 α in change of cellular size during senescence. Taken together, these data

suggest that each UPR signaling pathway plays an important role in the establishment of cellular senescence and that the observed features of cellular senescence depend on cell type, stimulus, and the activated signaling pathway(s). Although the role of ER stress in establishment of cellular senescence has been demonstrated, there are nearly no data on ER stress response and cellular senescence in lung aging. Since one of the major features of cellular senescence is the increased level of factors including interleukins, chemokines, growth factors, proteases and regulators, receptors, ligands, and ECM proteins (i.e., the SASP), senescent cells are most likely to be experiencing ER stress due to the increasing demand on protein folding. A direct consequence of ER stress is then the activation of UPR arms ATF6 and IRE1 α , which increase ER folding capacity and function (13). This results in ER enlarge-

FIGURE 3. Endoplasmic reticulum (ER) stress response induces cellular senescence

The ER stress triggering compounds including thapsigargin, tunicamycin, and dithiothreitol (DTT) induce cellular senescence through the activation of unfolded protein response (UPR) signaling pathways including inositol-requiring enzyme-1 (IRE1), protein kinase RNA-like ER kinase (PERK), and activating transcription factor-6 (ATF6). PERK signaling induces cell cycle arrest by inhibiting cyclin B1 and activating p21 through growth arrest and DNA-damage-inducible protein 45 α and 34 (GADD45 α and GADD34), respectively. ATF6 signaling controls the expression of senescence-associated secretory phenotype (SASP) molecule IL6, markers of cell cycle arrest including p53 and p21, senescence-associated β -galactosidase (SA- β -GAL) activity, and morphology change of senescent cells. In line with ATF6 signaling, IRE1 α signaling regulates the SA- β -GAL activity and IL6 synthesis. Additionally, the oxidative stress induces cellular senescence through activation of UPR arm ATF6, which promotes an increased expression of p21 and SA- β -GAL activity. COX2, cyclooxygenase 2.

ment of senescent cells (89). IRE1 α and PERK signaling pathways can also attenuate protein synthesis thereby reducing the folding demand, which in turn increases the ER folding capacity. Thus it is possible that IRE1 α and PERK signaling pathways may then prevent senescent cells being under severe ER stress and promote apoptosis. **FIGURE 3** illustrates the actual interaction between ER stress response and senescent pathways. However, these concepts investigating the ER stress response and cellular senescence cross talk have not been explored in the context of lung aging.

Conclusions and Future Challenges

In this review, we aimed to overview the current knowledge on ER stress response and cellular senescence cross talk in lung aging and highlight the fields that need to be investigated to improve our knowledge on the roles that these biological processes play in normal lung aging and pathogenesis of COPD. There are indications that ER stress response and cellular senescence play an important role in lung aging and COPD. Additionally, cell types including fibroblasts (parenchyma and airway), ASMCs, and epithelial cells have been less used for the characterization of ER stress response. To date, the available data on ER stress response and cellular senescence and their intercommunication are insufficient for a better understanding of their contributions to lung aging and COPD; these fields therefore need more investigations. Additionally, data highlighting sex-related differences in ER response in aged lung are not available. Thus the future challenges will be to 1) characterize the ER stress response and identify the natural inducers of ER stress in aged lung, 2) examine the cross talk between the ER stress response and senescence and identify the involved UPR arms, 3) examine the sex-related differences in ER stress in lung aging, and 4) investigate the role of ER stress response and senescence cross talk in directing ECM remodeling, as the role of ER stress response and cellular senescence have been suggested in pulmonary fibrosis. Elucidating these mechanisms might offer a new therapeutic strategy for COPD and IPF as diseases of aging in the lung. ■

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