Endoplasmic reticulum stress in alveolar epithelial cells is prominent in IPF: association with altered surfactant protein processing and herpesvirus infection

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Lawson WE, Crossno PF, Polosukhin VV, Roldan J, Cheng DS, Lane KB, Blackwell TR, Xu C, Markin C, Ware LB, Miller GG, Loyd JE, Blackwell TS. Endoplasmic reticulum stress in alveolar epithelial cells is prominent in IPF: association with altered surfactant protein processing and herpesvirus infection. Am J Physiol Lung Cell Mol Physiol 294: L1119–L1126, 2008. First published April 4, 2008; doi:10.1152/ajplung.00382.2007.—Recent evidence suggests that dysfunctional type II alveolar epithelial cells (AECs) contribute to the pathogenesis of idiopathic pulmonary fibrosis (IPF). Based on the hypothesis that disease-causing mutations in surfactant protein C (SFTPC) provide an important paradigm for studying IPF, we investigated a potential mechanism of AEC dysfunction suggested to result from mutant SFTPC expression: induction of endoplasmic reticulum (ER) stress and the unfolded protein response (UPR). We evaluated biopsies from 23 IPF patients (including 3 family members with L188Q SFTPC mutations, 10 individuals with familial interstitial pneumonia without SFTPC mutations, and 10 individuals with sporadic IPF) and sections from 10 control lungs. After demonstrating UPR activation in cultured A549 cells expressing mutant SFTPC, we identified prominent expression of UPR markers in AECs in the lungs of patients with SFTPC mutation-associated fibrosis. In individuals with familial interstitial pneumonia without SFTPC mutations and patients with sporadic IPF, we also found UPR activation selectively in AECs lining areas of fibrotic remodeling. Because herpesviruses are found frequently in IPF lungs and can induce ER stress, we investigated expression of viral proteins in lung biopsies. Herpesvirus protein expression was found in AECs from 15/23 IPF patients and colocalized with UPR markers in AECs from these patients, ER stress and UPR activation are found in the alveolar epithelium in patients with IPF and could contribute to disease progression. Activation of these pathways may result from altered surfactant protein processing or chronic herpesvirus infection.

idiopathic pulmonary fibrosis (IPF) is a chronic lung disease characterized by interstitial infiltrates in the lung parenchyma, restriction on pulmonary function testing, progressive dyspnea, and impaired exercise tolerance (8). Despite extensive research involving IPF and its pathologic correlate, usual interstitial pneumonia (UIP) (1), the critical factors for disease initiation and progression remain unknown. Recent investigations of familial interstitial pneumonia (FIP) have begun to shed light on important aspects of pathogenesis by identifying genetic mutations associated with disease (3, 12, 23, 27). We identified a mutation in the carboxy-terminal region of the gene encoding surfactant protein C (SFTPC) in a large FIP family, including 11 adults with UIP (27). This mutation is a heterozygous exon 5 +128 T-to-A transversion that substitutes glutamine for leucine at amino acid position 188 (L188Q) in the carboxy-terminal region of the precursor protein for surfactant protein C (pro-SP-C). Currently, several different mutations in SFTPC have been detected in adults and children with chronic interstitial lung disease (4, 18), localized primarily in the COOH-terminal BRICHOS domain.

BRICHOS domain proteins include the British and Danish dementia (BRI) family of proteins, CA11, chondromodulin-1 (ChM-1) and SP-C, which share a common COOH-terminal domain that is required for protein processing. Mutations in the BRICHOS domain lead to disease through mechanisms related to altered posttranslational protein processing and cell toxicity (4, 16). In SFTPC, BRICHOS domain mutations are predicted to affect pro-SP-C folding and disulfide-bond formation in the endoplasmic reticulum (ER) (4, 16). Overexpression of a prominent BRICHOS domain mutation of SFTPC (exon 4 deletion) in cultured epithelial cells leads to abnormal intracellular processing of pro-SP-C, ER stress, and activation of the unfolded protein response (UPR) (16). Accumulation of large amounts of protein in the ER results in activation of the UPR, which is designed to abrogate the effects of the misfolded protein. The UPR comprises pathways that globally attenuate protein translation, enhance expression of metabolism and redox proteins, increase production of folding chaperone proteins, and induce expression of protein degradation enzymes. While designed to attenuate ER stress, prolonged UPR activation can activate cellular apoptosis pathways (21).

In the mature lung, SP-C is expressed specifically by type II alveolar epithelial cells (AECs) (31). In our previous report, evaluation of the effects of L188Q SFTPC expression in AECs demonstrated evidence of aberrant surfactant processing and increased cytotoxicity in these cells (27). Therefore, we hy-
indicated that L188Q SFTPC alters the phenotype of AECs by inducing ER stress and activating the UPR. UPR activation in AECs could contribute to lung fibrosis by impairing cell survival and the ability to re-epithelialize air spaces after injury. If SFTPC mutations represent an important paradigm for disease in IPF, then activation of the UPR could be a common feature of IPF, both familial and sporadic. Although SFTPC mutations are rarely found in IPF (11), other factors that impact surfactant processing, cellular metabolism, or homeostasis could result in ER stress and UPR activation in type II AECs. For example, chronic herpesvirus [Epstein-Barr virus (EBV), cytomegalovirus (CMV), Kaposi’s sarcoma herpes virus (KSHV)] infection of the lung can be found in a high percentage of IPF subjects (25), and herpesvirus infection has been shown to induce ER stress in cell model systems (10).

To evaluate our hypothesis, we investigated whether expression of L188Q SFTPC results in ER stress and UPR activation in A549 cells and murine lung epithelial (MLE12) cells in vitro. We then evaluated lung biopsy specimens from FIP patients with and without SFTPC mutations and sporadic cases of UIP/IPF for evidence of ER stress and activation of the UPR by assessing expression of the following proteins in AECs: ER chaperone immunoglobulin heavy-chain-binding protein (BiP), X-box binding protein 1 (XBP-1), EDEM (ER degradation enhancing α-mannosidase-like protein), and phosphorylated eukaryotic initiation factor 2α (p-eIF2α) (14, 22). Our findings indicated that L188Q SFTPC expression results in ER stress and UPR activation in A549 and MLE12 cells in vitro and in type II AECs in vivo. In addition, ER stress and UPR activation was found in AECs from all patients with familial and sporadic IPF in the absence of SFTPC mutations. Interestingly, expression of herpesvirus proteins colocalize to this same cell population in the majority of patients. Together, these studies indicate that ER stress and UPR activation are present in AECs in IPF, may be influenced by hereditary as well as acquired factors, and could contribute to disease progression.

MATERIALS AND METHODS

Lung tissue sections. This investigation was approved by the Vanderbilt University Institutional Review Board, and informed consent was obtained. Lung samples were obtained from surgical lung biopsies performed for evaluation of interstitial lung disease or from explanted lung obtained at the time of lung transplantation. Lung biopsies were evaluated from 3 L188Q SFTPC family members with UIP, 10 individuals with UIP from non-SFTPC mutation FIP families, and 10 individuals with sporadic UIP. Diagnosis was made in accordance with American Thoracic Society/European Respiratory Society Consensus Statements (1, 2). Biopsies showing normal lung parenchyma obtained from patients undergoing lung nodule resection (three samples) or from explanted donor lungs that were deemed not suitable for transplantation (seven samples) were used as controls. Sequencing of SFTPC was performed on FIP individuals with biopsy proven UIP using methods previously reported (11). Cell culture studies. Expression constructs were generated containing wild-type human SFTPC (WT SP-C) and SFTPC with exon 5 + 128 T → A mutation (mutant L188Q SP-C) as previously described (27). Stably transfected A549 cells and MLE12 cells expressing WT and L188Q SP-C were obtained by G418 selection after transfection using Effectene (Qiagen, Valencia, CA). In pooled stably transfected cells, BiP levels were assessed on cell lysates by Western blot using a goat polyclonal antibody (Santa Cruz Biotechnology, Santa Cruz, CA) as previously described (13). Results were normalized to p44/p42 MAPK (antibodies from Cell Signaling Technology, Danvers, MA).

To determine the level of apoptosis, stably transfected cells were grown to confluence and then analyzed with the DeadEnd fluorometric terminal deoxynucleotidyltransferase-mediated dUTP nick end labeling (TUNEL) assay (Promega, Madison, WI).

Immunohistochemistry. Formalin or paraformaldehyde fixed paraffin embedded lung tissue was sectioned (5 μm), and immunostaining was performed using methods outlined previously (13). The primary antibodies listed below were used followed by a standard immunoperoxidase/avidin-biotin complex protocol (Vectastain ABC kit; Vector Laboratories, Burlingame, CA). Counterstain was performed with hematoxylin. For immunofluorescence, secondary FITC-conjugated and Texas red-conjugated antibodies (Jackson Immunoresearch, West Grove, PA) were used with methods outlined previously (13).

Primary antibodies used: BiP, also known as glucose regulated protein-78 (GRP78) - goat polyclonal antibody (Santa Cruz Biotechnology); XBP-1 - rabbit polyclonal antibody (Santa Cruz Biotechnology); EDEM - goat polyclonal antibody (Santa Cruz Biotechnology); eIF2α - rabbit polyclonal antibody (Santa Cruz Biotechnology); p-eIF2α - rabbit monoclonal antibody (Epitomics, Burlingame, CA); pro-SP-C - rabbit polyclonal antibody (Chemicon, Temecula, CA); EBV latent membrane protein 1 (EBV LMP1) - Dako CS 1–4 mixture of 4 mouse monoclonal antibodies (Dako, Carpinteria, CA); CMV - mouse monoclonal antibodies to late antigens clones 1G5.2 and 2D4.2 (Chemicon) and immediate early and early antigens clones CCH2 and DDG9 (Dako); KSHV K-cyclin - sheep polyclonal antibody (Exalpha Biologicals, Maynard, MA). For each batch of samples, negative controls were performed in which no primary antibodies were added.

Light microscopy was performed with an Olympus IX70 microscope (Olympus Optical, Tokyo, Japan), and images were captured with an Optronics digital camera and MagnaFire software (Optronics, Goleta, CA). Immunofluorescence imaging was performed using an LSM-510 META Laser Scanning Microscope (Carl Zeiss Advanced Imaging Microscopy, Jena, Germany) with Z-stack images obtained.

Semi-quantitative assessment of immunohistochemistry. Slides immunostained for BiP, EDEM, and XBP-1 were reviewed by a lung pathologist blinded regarding specimen source. Each specimen was scored for protein expression in AECs on a 0–3 scale, with 0 = no positive cells, 1 = positive immunostained cells comprising <25% of the epithelial cells on the section, 2 = 25–50% of the epithelial cells on the section, 2 = 25–50% of the epithelial cells on the section, 3 = >50% of the epithelial cells on the section were immunostain positive. For herpesvirus proteins, immunohistochemistry (IHC) studies were determined as positive or negative for each specimen depending on the presence or absence of immunostaining in the alveolar epithelium.

Statistics. Statistical analyses were performed using GraphPad Instat (GraphPad Software, San Diego, CA). To assess differences among groups for the TUNEL assay, an one-way analysis of variance test was performed. To evaluate differences between IPF and normal lung tissue with respect to UPR marker expression or presence of herpesviruses, a Fisher’s exact test was used. Results are presented as means ± SE. P values <0.05 were considered significant.

RESULTS

Mutant L188Q SFTPC causes ER stress and UPR activation. To determine whether expression of L188Q mutant pro-SP-C induces ER stress in AECs, we stably transfected A549 cells with vectors expressing either WT or L188Q SFTPC. We then evaluated the expression of BiP, a well-recognized marker of ER stress (14) by Western blot (Fig. 1). As shown, WT pro-SP-C expression resulted in increased BiP expression compared with untransfected A549 cells; however, BiP levels were substantially higher in cells expressing L188Q pro-SP-C, indicating the presence of ER stress in these cells. Recently, Mulugeta et al. (15) found similar findings by transient transfection of A549 cells with a plasmid expressing the L188Q
SFTPC construct, demonstrating evidence of ER stress and caspase pathway activation. In separate experiments, we analyzed MLE12 cells stably transfected with the WT or L188Q SFTPC constructs for apoptosis. By fluorimetric TUNEL assay, MLE12 cells transfected with L188Q SFTPC had a greater number of apoptotic cells per high power field than did untransfected or WT SFTPC transfected cells (Fig. 2).

In UIP, areas of fibrotic remodeling contain air spaces lined by hyperplastic type II AECs. To demonstrate pro-SP-C expression by these type II AECs, we performed IHC for pro-SP-C on lung tissue from individuals with UIP in the presence or absence of an L188Q SFTPC mutation. As shown in Fig. 3, AECs lining areas of fibrosis uniformly express pro-SP-C.

Next we analyzed markers of ER stress and UPR activation (BiP, EDEM, XBP-1, and p-eIF2α) in lungs of three individuals with biopsy proven UIP from the previously reported L188Q SFTPC mutation family (27) and 10 control lung sections from individuals without IPF (Fig. 4). Immunostaining for BiP, EDEM, and XBP-1 revealed prominent staining in epithelial cells lining areas of affected lung in the patients with UIP from the L188Q SFTPC family. In contrast, control lungs with normal histological appearance were negative for BiP, EDEM, and XBP-1. Interestingly, 6/10 of the control sections from donor lungs that were rejected for transplantation had evidence of inflammation and tissue edema, likely contributing to their rejection for use in transplantation. These sections exhibited low-level BiP staining, indicating that diseased controls may have increased BiP expression; however, EDEM and XBP-1 staining were not identified in any of the control lung sections. While widespread eIF2α staining was identified in both normal and fibrotic lungs, the phosphorylated form that is associated with the UPR (p-eIF2α) was only detected in very rare epithelial cells in UIP biopsies (not shown), suggesting that there is some selectivity in the UPR induced by ER stress in AECs in vivo. Together, these data show that expression of L188Q pro-SP-C in vitro and in vivo results in ER stress and activation of the UPR. These findings are consistent with the idea that SFTPC mutations result in a protein product that cannot be folded and processed properly, leading to ER stress.

ER stress and UPR activation are found in familial and sporadic IPF without SFTPC mutations. On the basis that the hypothesis that the pathogenesis of familial and sporadic IPF is similar and that SFTPC mutations provide an important disease
paradigm, we evaluated lung sections from an additional 10 patients with FIP in the absence of an SFTPC mutation and 10 patients with sporadic IPF for the presence of ER stress and UPR markers. In all patients, the pathologic diagnosis was UIP. IHC studies for BiP, EDEM, and XBP-1 on formalin fixed tissue from FIP and sporadic IPF patients indicated that the distribution of staining was similar to that observed in lung biopsies from patients with L188Q SFTPC mutations (Fig. 4). Immunostaining of AECs in areas of fibrosis was identified for all three markers, whereas minimal staining was observed in other cell types in the lungs. The results of these studies indicate that the UPR is specifically activated in epithelial cells lining areas of affected lung in IPF. A few of the lung biopsies from IPF patients contained areas with normal-appearing lung architecture, and in these areas, some type II AECs were immunopositive for ER stress markers. As with the tissue specimens from patients with L188Q SFTPC mutations, widespread eIF2α expression was detected in all lung specimens, but p-eIF2α was not detected in the lining epithelial cells.

Semi-quantitative scoring (on a 0–3 scale) of the IHC studies for BiP, EDEM, and XBP-1 was done to evaluate the extent of UPR activation in AECs in areas of fibrotic remodeling. As shown in Fig. 5, >50% of AECs in lung sections from the patients with L188Q SFTPC mutations stained positive for all three UPR markers (score of 3). High BiP scores (2 or 3) were found in all but one of the FIP and sporadic IPF cases. Low-level BiP staining (score of 1) was noted in 6/10 control lung sections but not detected in the other four control lung sections. In addition to BiP, EDEM- and XBP-1-positive AECs were present in all FIP and sporadic IPF cases, but not in any of the control lung specimens. For statistical analysis, all biopsies from UIP were compared with controls. For BiP staining, 23/23 IPF lung sections were immunostain positive compared with 6/10 controls (Fisher’s exact test, \( P < 0.005 \)). For both EDEM and XBP-1 staining, 23/23 IPF lung sections were immunostain positive compared with 0/10 controls (Fisher’s exact test, \( P < 0.0001 \)). Together, these data indicate that AECs in FIP associated with L188Q SFTPC mutations and AECs in FIP and sporadic IPF in the absence of SFTPC mutations share common pathways involving ER stress and UPR activation.

Herpesvirus proteins are detected in AECs with ER stress. Since we detected evidence of widespread UPR activation in fibrotic lungs in the absence of SFTPC mutations, we considered other factors that could induce ER stress in AECs. Herpesvirus antigens are present in the lungs of a majority of UIP/IPF patients (25), with prior studies showing localization of EBV LMP1 to the lung epithelium in IPF (28). Furthermore, herpesviruses have been shown to induce ER stress in vitro (10). Therefore, we investigated whether herpesvirus antigens were present in AECs in IPF patients and might contribute to ER stress and UPR activation in these cells. Using IHC...
techniques on sections serial to the ones used to identify UPR markers, we detected evidence of infection by EBV, CMV, and KSHV in the lungs of the IPF patients in which we had identified UPR activation. Herpesvirus antigens were identified in the alveolar epithelium in 2/3 L188Q SFTPC cases, 6/10 familial cases, 7/10 sporadic cases, but in 0/10 control lung cases. Thus, herpesviruses were detected in 15/23 from the combined UIP group versus 0/10 in the controls (Fisher’s exact test, \(P = 0.0005\)). In each of these categories of IPF patients, one individual had evidence of two herpesviruses. Table 1 summarizes these results. The viral antigens detected by IHC in these studies are found in chronic herpesvirus infection, as demonstrated in the CMV-positive cases. Late CMV antigens were detected, while immediate early and early CMV antigens were not identified (data not shown). Examples of immunohistochemistry studies for CMV, KSHV, and EBV are shown in Fig. 6, with serial sections demonstrating that herpesvirus proteins were found in the alveolar epithelium lining areas of fibrosis in the same distribution seen for the ER stress markers described above (Fig. 6, C and D). To expand on the findings noted with serial section IHC studies, dual fluorescence microscopy studies were performed in IPF lung sections. In those samples that were herpesvirus positive, UPR markers and herpesvirus proteins were found to colocalize to the same AEC population. Most of the AECs that were herpesvirus positive were also positive for the UPR marker (BiP or XBP-1). An illustrative example of these studies is shown in Fig. 7, where XBP-1 and late CMV antigens are found to colocalize to the AECs lining an area of fibrosis. These studies suggest that herpesvirus infection may contribute to the ER stress and UPR activation present in type II AECs in patients with IPF.

**DISCUSSION**

In this study, we have shown that expression of L188Q SFTPC results in ER stress and UPR activation. We then provided evidence that ER stress and UPR activation are present in AECs lining areas of affected lung in IPF, whether or not an SFTPC mutation is present. The similarities seen in fibrosis in the same distribution seen for the ER stress markers described above (Fig. 6, C and D). To expand on the findings noted with serial section IHC studies, dual fluorescence microscopy studies were performed in IPF lung sections. In those samples that were herpesvirus positive, UPR markers and herpesvirus proteins were found to colocalize to the same AEC population. Most of the AECs that were herpesvirus positive were also positive for the UPR marker (BiP or XBP-1). An illustrative example of these studies is shown in Fig. 7, where XBP-1 and late CMV antigens are found to colocalize to the AECs lining an area of fibrosis. These studies suggest that herpesvirus infection may contribute to the ER stress and UPR activation present in type II AECs in patients with IPF.

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this study between SFTPC mutation- and non-SFTPC mutation-associated lung fibrosis suggest common pathways in disease pathogenesis, specifically that ER stress and UPR activation may be important contributing factors to the disease process in IPF. Although altered surfactant processing may contribute to ER stress in epithelial cells in the lungs of IPF patients, colocalization of herpesvirus antigens and markers of the UPR in AECs in a large percentage of IPF patients suggests that herpesvirus infection could also contribute to ER stress in these cells.

With ER stress, UPR pathways are activated in an initial attempt to protect the cell (21). The UPR involves three ER transmembrane proteins that act as sensors for ER stress, PKR-like ER kinase (PERK), activating transcription factor 6 (ATF6), and inositol-requiring enzyme 1 (IRE-1) (10, 21). Under normal conditions, these sensors are bound by BiP and maintained in an inactive state. When misfolded proteins accumulate in the ER, BiP is released to serve as a folding chaperone (5), allowing each sensor to assume its active state. PERK activation results in global attenuation of protein translation through a p-eIF2α-dependent mechanism and in an ATF4-dependent expression of metabolism and redox proteins such as GADD34 (growth arrest and DNA damage inducible gene 34) designed to protect the cell (9). Subsequently, translational recovery is mediated through the ability of GADD34 to dephosphorylate eIF2α (19), which may explain the relative lack of detection of p-eIF2α by IHC in our studies. ATF6 activation results in the expression of chaperone proteins, including BiP, which target further attempts to assist in protein folding (30) and in the increased expression of XBP-1 (32). The intrinsic endonuclease activity of IRE-1 results in the splicing of XBP-1 to its active state leading to expression of chaperones and UPR-mediated protein degradation enzymes, including EDEM (32). While designed to attenuate ER stress, severe or prolonged UPR activation can activate cellular apoptosis pathways including ER bound caspase-4 (17, 21).

The downstream mechanism by which induction of ER stress and UPR activation contribute to IPF is not yet clear; however, multiple studies have implicated apoptosis of AECs in the pathogenesis of lung fibrosis (26). Thus, it is possible that ER stress directly induces apoptosis of AECs or contributes to apoptosis following exposure to toxic or injurious stimuli, leading to an inability to re-epithelialize areas of injured lung parenchyma. Activation of the UPR could also impact other important functions of type II AECs, like surfactant production, that disrupt homeostasis in the lung parenchyma and contribute to fibrosis.

If ER stress is an important force behind the ongoing injury and aberrant repair pattern seen in IPF, defining the etiology of ER stress in AECs and identifying approaches to ameliorate ER stress may be important future directions for research in interstitial lung disease. Since mutations in SFTPC are rarely seen in FIP or sporadic IPF, it is unlikely that inherited defects in pro-SP-C are responsible for the widespread ER stress and UPR activation identified in IPF samples. However, other inherited or acquired defects in proteins processed by the secretory pathway of type II AECs or dysfunctional proteins critical to the operation of this pathway could result in inefficient protein processing and ER stress. Our studies focused on a potential role for herpesvirus in ER stress and activation of the UPR. Previous studies have demonstrated evidence of EBV in IPF lungs by both PCR for viral DNA and IHC for viral proteins (7, 24, 28). In 2003, Tang et al. (25) analyzed all known herpesviruses by DNA PCR and found that EBV, CMV, and KSHV (HHV-8) were present significantly more frequently in IPF lungs than controls. More than 90% of IPF patients were found to be PCR positive for herpesviruses in the lungs, whereas two other viruses reactivated by immunosuppression were not detected (20). As with misfolded proteins, herpesvirus infections can activate the UPR because of viral proteins produced and passed through the ER (6, 10, 29). Remarkably, however, the normal consequences of UPR activation are modified to benefit the virus. For example, in CMV infection, UPR pathways are activated but translation attenuation and expression of degradation factors do not occur (10). Interestingly, we did not find increased p-eIF2α, the UPR component associated with global attenuation of protein translation, in the lungs of IPF patients despite the presence of other markers of UPR activation.

We demonstrated that herpesvirus proteins (CMV, EBV, and KSHV) can be detected in the same AECs that show evidence of ER stress and UPR activation, suggesting that herpesviruses play a role in IPF disease progression through induction of this pathway. Although this is an intriguing possibility, more work is required to determine the timing and role of herpesvirus infections in IPF. Not all individuals we studied with familial or sporadic IPF in the absence of SFTPC mutations had...
evidence of herpesviruses protein expression in the lungs, arguing that other factors besides herpesvirus infection and mutant STT4C expression can induce ER stress in AECs from patients with IPF. However, we did not study all possible herpesviruses or utilize PCR techniques, which could be more sensitive than IHC, for identification of herpesviruses.

With these studies, we have shown that ER stress and UPR activation are found in specific cell populations in the lungs of individuals with FIP and sporadic IPF. Although we suggest that UPR activation could contribute to fibrotic remodeling in IPF, these studies do not prove causality and other explanations for our findings are possible. For example, ER stress and UPR activation in the hyperplastic type II AEC population could be an adaptive response to increased protein production as these cells attempt to re-epithelialize areas of injured lung. Delineating a true cause and effect relationship between ER stress and subsequent pulmonary fibrosis as well as the contribution of herpesviruses to this process will require further study.

While the causative factors in IPF remain largely undefined, the finding that STT4C mutations can cause UIP in families highlights the importance of the type II AEC in the disease process. We have extended these observations regarding type II AECs in IPF by providing evidence that ER stress and UPR activation in AECs are also found in IPF. Furthermore, we propose that this pathway is central to disease pathogenesis and that delineating the cause of ER stress and the subsequent response of type II AECs, as well as further analysis of the role of herpesviruses in this process, should be targets of future investigations.

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ER STRESS IN IPF


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