

Editorial

Outstaying their Welcome: The Persistent Myofibroblast
in IPF

Steven K. Huang and Jeffrey C. Horowitz*

Division of Pulmonary and Critical Care Medicine,
Department of Internal Medicine, University of Michigan
Medical Center, USA***Corresponding author:** Jeffrey C. Horowitz, Division
of Pulmonary and Critical Care Medicine, Department
of Internal Medicine, University of Michigan Medical
Center, 6301 MSRB III, 1150 W. Medical Center Drive,
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Pulmonary fibrosis is the final common end point of a variety of acute and chronic causes of lung injury. Regardless of the cause, it represents an aberrant wound-repair response characterized by the excessive accumulation of extracellular matrix (ECM) and the replacement of normal lung parenchyma with scar tissue. Fibroblasts are mesenchymal cells that serve a critical role in both normal and fibrotic repair processes. These cells exist along a spectrum of activation states spanning from relatively undifferentiated quiescence during times of homeostasis to differentiated, highly secretory and contractile smooth muscle-like cells with organized α -smooth muscle actin-containing stress fibers (i. e. myofibroblasts) following tissue injury [1]. In their fully activated state, myofibroblasts are the primary cells responsible for the synthesis, secretion, and remodeling of the ECM. While the resolution of normal wound repair coincides with substantial myofibroblast apoptosis, fibrotic repair is associated with the continued accumulation of these activated cells [2,3].

Idiopathic pulmonary fibrosis (IPF) is the most common and clinically refractory of the idiopathic interstitial lung diseases. Our understanding of IPF pathogenesis has evolved from one in which fibrosis was driven by excessive inflammation to one in which fibrosis results from dysfunctional interactions between an injured epithelium, reparative fibroblasts and select inflammatory cells [4,5]. Specifically, a role for alternatively activated (M2) macrophages and/or fibrocytes, has been recognized [6,7]. Although this understanding continues to evolve, specific pathologic lesions called “fibroblastic foci” have been consistently associated with IPF and provide a window into the pathobiology of IPF [8]. These foci are comprised of activated myofibroblasts in close approximation to an injured alveolar epithelium. While not unique to IPF, these foci are thought to represent the site of “active” fibrosis within IPF lungs. Clinically, the number of fibroblastic foci correlates with mortality in IPF [9]. Biologically, these foci are characterized by an “apoptosis paradox” wherein there is prominent epithelial cell apoptosis but insufficient mesenchymal cell apoptosis [3,10]. The aberrant interactions and dysfunctional cross-talk between fibroblasts and epithelial cells may promote a self-perpetuating cycle that maintains this apoptosis paradox, even if the initial stimulus of epithelial injury has abated [8].

As noted, the unrestrained accumulation of myofibroblasts is a key feature that differentiates fibrotic from physiologic repair. The accrual of these cells represents the combined effects of cell trafficking, proliferation and death. Of these, proliferation has received considerable attention; indeed, fibroblastic foci were initially defined as “small aggregates of actively proliferating fibroblasts and myofibroblasts” [11,12]. Certainly, studies suggest that fibroblast proliferation is important, especially in the early phases of wound repair and fibrosis [13]. Moreover, soluble mediators implicated in fibrosis have been shown to induce fibroblast proliferation in vitro [14-16]. Comparisons of IPF and normal lung fibroblasts, however, have shown variable results, and some studies suggest that IPF fibroblasts actually have a decreased rate of proliferation [17,18]. Moreover, studies of IPF tissue have demonstrated that while there is substantial epithelial cell proliferation, there is little to suggest robust fibroblast proliferation within fibroblastic foci [19,20].

Decreased fibroblast apoptosis within the fibroblastic foci represents another mechanism by which these cells accumulate. In support of this, studies of IPF lung biopsies have consistently shown a lack of apoptotic cells within the myofibroblast niche [19-24], and a growing body of literature supports the hypothesis that the acquisition of an apoptosis-resistant phenotype contributes to the fibroblast accumulation in IPF [22,23,25-29]. The mechanisms underlying apoptosis resistance are likely multifactorial and may contribute to the clinical heterogeneity observed in patients with IPF. Soluble mediators strongly associated with fibrosis, most notably transforming growth factor beta-1 (TGF- β 1) and endothelin-1 (ET-1), promote fibroblast resistance to death receptor-mediated apoptosis via activation of focal adhesion kinase (FAK) and the PI3K/AKT signaling pathways [30-33]. Each of these pro-survival protein kinases has been found to be expressed at increased levels in IPF fibroblasts and/or fibroblastic foci, and inhibition of these kinases has been shown to attenuate lung fibrogenesis in animal models [34-39]. TGF- β 1 and ET-1 also utilize FAK and AKT to induce expression of endogenous inhibitors of apoptosis, including X-linked inhibitor of apoptosis (XIAP) and survivin, which are expressed at increased levels in fibrotic lung fibroblasts and in the fibroblastic foci of IPF tissue [22,26,33]. Pharmacologic blockade or gene-silencing of these and other endogenous inhibitors of apoptosis enhances fibroblast sensitivity to apoptotic stimuli [26,27,29,32,33]. In contrast, antifibrotic mediators, such as prostaglandin E₂ (PGE₂), induce fibroblast apoptosis through decreased AKT signaling and suppression of XIAP and survivin [22,40]. Diminished PGE₂ production and/or responsiveness may, therefore, also contribute to fibroblast apoptosis resistance [41]. Several studies have also shown decreased expression of Fas/CD95, a receptor which mediates apoptosis induced by extrinsic signals such as Fas ligand [23,42]. Loss of Fas/CD95 expression is mediated, in part, through epigenetic mechanisms [43]. Stimuli that increase Fas/CD95 expression on fibroblasts lead to their increased susceptibility to apoptosis [44].

Beyond the realm of soluble mediators, fibroblast interactions with the ECM are critical in the regulation of fibroblast survival and apoptosis. Disruption of biochemical and biomechanical signals generated by cell-matrix interactions can enhance sensitivity to apoptosis [45-47]. Our studies have shown that plasminogen activation, which is strongly linked with protection from fibrosis in murine models, induces fibroblast apoptosis in conjunction with fibronectin proteolysis [45]. In addition, recent studies show that alterations in ECM stiffness is sufficient to modulate fibroblast apoptosis and that fibroblasts have increased rates of apoptosis (and decreased proliferation) when exposed to substrates with a compliance similar to that of normal lung. As substrate stiffness increases toward that of fibrotic lung tissue, there is a decline in fibroblast apoptosis coupled with an increase in fibroblast proliferation [46]. These and other studies [48] suggest that the fibrotic lung matrix itself might be sufficient to support persistent myofibroblast survival and accumulation, even in the absence of ongoing lung injury or continued stimulation by soluble mediators.

IPF is a heterogeneous disease with dismal outcomes and inadequate treatment options [4,25]. The pathogenesis involves complex interactions between multiple cell types, multiple soluble mediators and dynamic cell-matrix interactions that result in the accumulation and persistence of fibroblasts and myofibroblasts. Further elucidation of the mechanisms regulating fibroblast proliferation and survival may lead to novel therapeutic interventions in IPF [25].

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