Notch3: A New Culprit in Fibrotic Lung Disease

The respiratory system is designed for efficient gas exchange in the distal region of the lung. It consists of conducting airways that include the trachea, bronchi, and bronchioles that link to distal alveoli. Multiple pathways have been identified that play a pivotal role in the maintenance of progenitor-cell populations and in rebuilding damaged lungs deep within the parenchyma. In this regard, the Notch pathway has been studied extensively with respect to its role in lung development and regeneration and has been found to play a role specifically in proximal–distal patterning and cell differentiation, proliferation, and apoptosis (1). Notch signaling is a highly conserved pathway that mediates cell–cell contacts; in an ideal setting, it facilitates communication between cells within a short radius of the activated cell. The canonical pathway consists of four receptors (Notch 1, 2, 3, and 4) and five ligands (Jagged 1 and 2, Dll 1 (Delta-like ligand 1), Dll 3, and Dll 4), which are membrane-bound on adjacent cells. Upon ligand binding, the NICD (Notch intracellular domain) of the receptor is cleaved by the γ-secretase enzyme and is translocated to the nucleus, where it

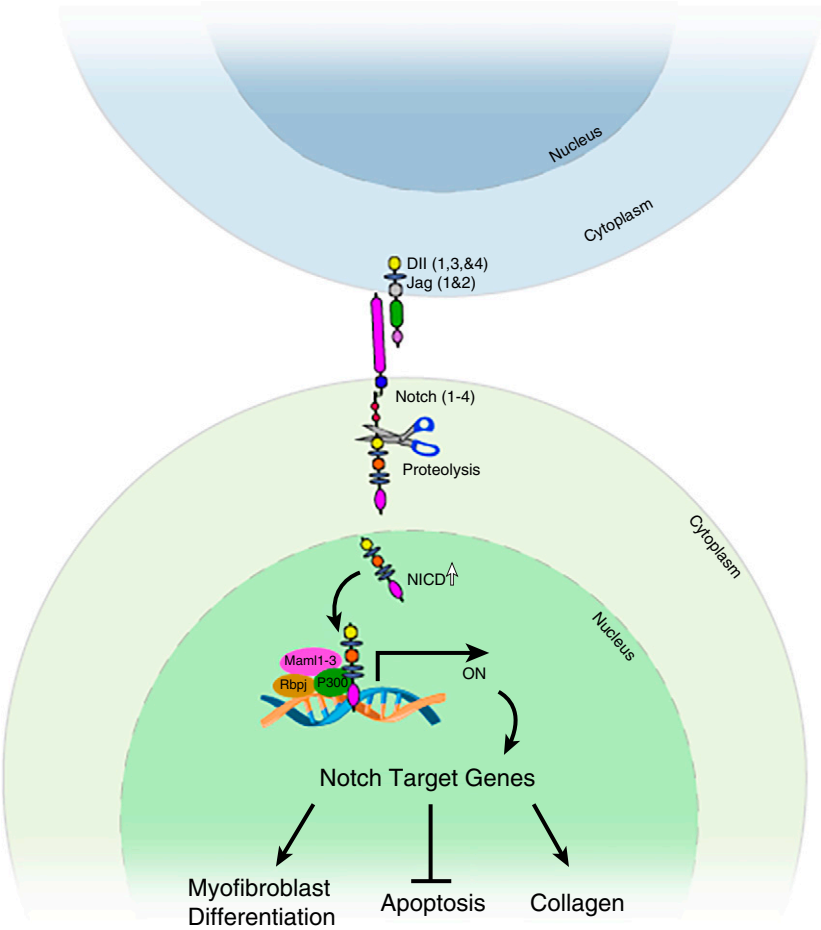

Figure 1. A general scheme illustrating the potential mechanistic pathway of Notch signaling in the pathogenesis of pulmonary fibrosis. The current study demonstrates that Notch3 is upregulated in fibroblasts and promotes myofibroblast transformation and survival, which is involved in excessive collagen deposition in the lung parenchyma. Black arrows highlight positive regulation. Perpendicular line highlights negative regulation. Dll = Delta-like ligand; Jag = Jagged ligand; Maml = mastermind-like protein; NICD = the intracellular domain of the Notch protein; ON = transcriptional activation; P300 = histone acetyltransferase p300; Rbp = recombination signal binding protein.

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acts as a transcription factor (Figure 1). There, it interacts with other inactive transcriptional complexes (Rbpj and Maml1 [mastermind-like 1], Maml2, and Maml3) and triggers the expression of Notch-mediated genes such as Hes (Hes family BHLH transcription factor 1) and Hey (2, 3). The Notch signaling pathway is known to play a prominent role during lung development, in maintaining homeostasis, responding to injury, and regeneration. During development, the Notch pathway is involved in proximal–distal patterning, cell-fate decisions in the conducting airways, and alveolar and vascular development (4). As Notch signaling plays a pivotal role in homeostasis and regeneration, dysregulation in Notch signaling can lead to respiratory disorders including lung cancer, pulmonary arterial hypertension, chronic obstructive pulmonary disease, and idiopathic pulmonary fibrosis (IPF) (5–9). IPF is a fatal fibrotic lung disease marked by key cellular events such as fibroblast proliferation, migration, myofibroblast differentiation, and resistance to apoptosis (10, 11). The lack of effective medical treatment to reduce mortality in IPF highlights the importance of identifying novel therapeutic targets that may be useful in preventing or delaying the progression of pulmonary fibrosis.

In this issue of the *Journal*, Vera and colleagues (pp. 465–476) describe studies uncovering a pathogenic role for Notch3 in fibroblast activation and pulmonary fibrosis (12). They demonstrate that Notch3 amounts are elevated during bleomycin-induced pulmonary fibrosis and that active forms of Notch3 are coexpressed by αSMA-positive myofibroblasts in both mice and humans. They employed Notch3 knockout mice to delineate the role of Notch3 in pulmonary fibrosis. In the absence of Notch3, the lungs were protected from bleomycin-induced pulmonary fibrosis, and very few myofibroblasts were observed in comparison with control fibrotic lungs. In addition, they show that Notch3 deficiency in fibrotic fibroblasts leads to apoptotic events *in vitro*; this suggests Notch3 has a role in the pathogenesis of IPF. In another independent study, elevated levels of Notch1 were identified in IPF tissues that promoted myofibroblast transformation (13). The importance of the Notch pathway in myofibroblast differentiation was shown in a study by Xu and colleagues in which the canonical Notch pathway was disrupted. After this disruption, the expression of αSMA in mesenchymal cells, which is required for alveogenesis during lung development, was inhibited (14). Similarly, *in vitro* and *in vivo* studies have suggested that Notch signaling augments the differentiation of fibroblasts to myofibroblasts (15). Notably, mesenchymal-specific deletion of Notch1 attenuated pulmonary fibrosis *in vivo* that was associated with decreased myofibroblast transformation and a decrease in collagen production in the lungs (5). Likewise, in response to injury, Notch3 levels were shown to increase in $PDGFR\beta^{+}$ fibroblasts that are distinct from pericytes, as shown by single-cell analysis (16). However, more preclinical studies should be performed to delineate the mechanisms underlying the upregulation of the active form of Notch receptors, and their target genes, with respect to myofibroblast differentiation. Nevertheless, a growing body of evidence has emerged suggesting that a Notch signaling cascade, including Notch 1 and 3, positively regulates fibroblast activation and pulmonary fibrosis *in vivo* (Figure 1). Thus, the members of the Notch signaling pathway are legitimate therapeutic targets that, when blocked, may inhibit fibroblast activation and pulmonary fibrosis. ■

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