Pumping the Brakes on Pulmonary Fibrosis: A New Role for Regulator of Cell Cycle

As a result of injury or damage, tissue fibrosis can occur in any organ throughout the body. Within the lungs, there is a robust repair response initiated through a variety of different physiological mechanisms. Although the wound healing response is necessary to preserve lung function, the repair process can go awry, resulting in fibrosis, which is defined as the accumulation of excessive extracellular matrix protein or fibrous connective tissue. The most severe form is termed idiopathic pulmonary fibrosis (IPF), which causes shortness of breath, respiratory distress, and ultimately death. There are several U.S. Food and Drug Administration–approved therapies for pulmonary fibrosis that delay disease progression, but currently there is no cure. One reason for the lack of therapeutic options is that fibrosis is a multicellular event that begins with injury to the epithelial cells, leads to recruitment and/or stimulation of the immune system, and results in the activation and differentiation of fibroblasts (1). Because of the complexity of these signaling pathways and cell-to-cell interactions, identification and characterization of molecular brakes are needed to halt or possibly reverse the damage caused by excessive collagen secretion and remodeling. The development of novel strategies to restore these braking mechanisms may halt the disease progression and allow the lung time to repair and reverse the disease process.

In this issue of the *Journal*, Luzina and colleagues (pp. 146–157) report that the protein RGCC (regulator of cell cycle) is a novel suppressor of fibrotic signaling in fibroblasts (2). The authors demonstrate that expression of RGCC in whole lung tissue isolated from both murine models of pulmonary fibrosis and patients with IPF was decreased when compared with normal, donor controls. Although RGCC is expressed in various cells throughout the lung, the decrease in fibroblasts was most pronounced, leading the investigators to focus on the role of this protein in that cell population. Interestingly, mice deficient in RGCC had paradoxical reactions to bleomycin-induced pulmonary fibrosis with reduced collagen production when challenged with one dose of bleomycin and elevated collagen when challenged with multiple doses, inducing a chronic fibrotic model. Using primary human lung fibroblasts, the authors show that overexpression of RGCC attenuates TGF-β (transforming growth factor-β) activation of fibroblasts by inhibiting Smad signaling. This study confirms that fibrosis is associated with diminished RGCC expression and broad, diminished antifibrotic effects in fibroblasts.

Although originally identified within the central nervous system, RGCC is abundantly expressed throughout the body and has a role in many fundamental biological processes, including cell proliferation and differentiation, tumorigenesis, innate and adaptive immunity, and fibrosis (3). The first description of RGCC in fibrosis was in a murine model of kidney injury and repair (4). In this model, RGCC was elevated early after injury and inhibition by shRNA resulted in reduced collagen deposition and α-SMA (α-smooth muscle actin expression), as a marker of fibroblast activation. More recently, investigators examined the role of RGCC in skin fibrosis and inflammation using a bleomycin-induced systemic sclerosis murine model (5). RGCC-deficient mice had reduced skin thickening, fibrosis, and decreased collagen deposition, as well as diminished macrophage infiltration and proinflammatory cytokine production. Interestingly, other reports have demonstrated that RGCC expression is dependent on the response to both profibrotic factors, such as TGF-β, and proinflammatory cytokines, such as TNF-α (6). Given the role of this pleotropic protein in regulating the cell cycle, inflammatory signaling, and fibrotic pathways, additional work is needed to investigate RGCC in injury and repair responses in different tissues.

To extend the current study and further assess the role of RGCC in pulmonary fibrosis, it is important to consider the issue of fibroblast heterogeneity within the lung microenvironment. Recent studies using animal models have demonstrated the presence of fibroblast subtypes across a variety of different organs (7). Within the human lung, single-cell RNA sequencing analysis of lung tissue from IPF or normal donor controls has identified distinct fibroblast subpopulations (8, 9). In this study, the authors examine RGCC expression using both human and animal models. Although whole lung samples from the murine models consistently demonstrate diminished RGCC expression after bleomycin challenge, human lung fibroblasts exhibit considerably more heterogeneity in their protein expression of RCGG. These issues could be related to differences in RGCC expression between species (mouse vs. human), types of samples (whole lung vs. isolated cells), or fibroblast subtypes. The question going forward is to how to align expression and functional data of specific proteins with the possibility of fibroblast heterogeneity within the lung microenvironment.

Given the high clinical significance of myofibroblast dedifferentiation studies, additional work evaluating the effect of RGCC in Smad-independent pathways would be worth pursuing. In a recent study published by Fortier and colleagues, RNA sequencing analysis revealed that RGCC was upregulated by dedifferentiation agents, such as prostaglandin E_2 and fibroblast growth factor 2, indicating a likely role for this protein in mediating the dedifferentiation process (10). Therefore, RGCC induction responses to dedifferentiation agents and the ability to signal through RGCC-dependent pathways are needed to better understand this protein's pleiotropic actions. Another characteristic feature of fibrotic fibroblasts is their notorious resistance to apoptosis (11). Although RGCC has not been studied extensively in this context, one study reported that loss of RGCC promotes cell survival in nerve cells (12), suggesting a possible role for this protein in apoptosis resistance.

Taken together, these results provide a strong foundation suggesting that the loss of RGCC in fibroblasts contributes to

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pulmonary fibrosis. Using murine models of pulmonary fibrosis and primary human lung fibroblasts isolated from patients with and without fibrosis, the authors demonstrate that RGCC regulates expression of collagen and α-SMA as well as extracellular collagen deposition. Additional studies are necessary to explore the mechanism by which RGCC regulates fibrotic signaling pathways. These studies provide an important launching point to investigate further biological effects of this protein in regulating cell proliferation and proinflammatory and profibrotic pathways. ■

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