Escalating Mucus Inhibition to the Top of Our Priorities

In the past 15 years we have come to understand how an airway epithelial secretory cell can be induced to become a factory for mucus production (1, 2). To maintain lung function during the stress of an infection or exposure, inflammatory cytokines modulate the respiratory epithelium to produce mucus to eliminate pathogens and cellular debris. Under basal conditions, goblet cells comprise one in 20 cells on the epithelial surface, and after an infection this may increase 10-fold (3, 4). After such an insult, the epithelium recovers over days to weeks and the lining cells revert to normal. Yet, in airway diseases, goblet cells persist and may constitute a majority of airway lining cells. This epithelial transformation contributes to mucoid impaction, airflow obstruction, and increased susceptibility to infection (5, 6).

Goblet cell development requires two signals: activators of EGFR (epidermal growth factor receptor) to inhibit epithelial cell apoptosis, and IL-13 to activate STAT6 (signal transducer and activator of transcription 6). STAT6 activation stimulates goblet cell secretory functions through effects on transcription factors, including FOXM1 (forkhead box M1) and SPDEF (SAM pointed domain containing ETS transcription factor). These factors induce mucin gene expression and induction of $GABA_A$ receptors that enhance airway epithelial cell proliferation and further increase mucin production and secretion (7, 8). The initial signals through EGFR and STAT6 turn on numerous genes involved in the machinery for mucus production and secretion, and they also turn down repressors of goblet cell development such as FOXA2 and TTF1 (thyroid transcription factor) (2, 9–12). The hierarchy of factors and their effects has been elegantly detailed in mouse models in which individual genes in the pathway were mutated or blocked, and these findings were later confirmed in human airway epithelial cells in culture (8, 13).

In this issue of the *Journal*, Feldman and colleagues (pp. 322–331) add another layer to the complex interplay of signals that regulate goblet cell differentiation (14). The authors show that phospho-SMAD signaling is one of the principal pathways restricting goblet cell differentiation. Phospho-SMAD is high in basal and secretory cell precursors and low in goblet cells, and in response to SMAD inhibitors, IL-13–induced goblet cell development and mucin production are increased. Thus, SMAD signaling appears to be an important gatekeeper to limit goblet cell differentiation, and the authors were able to place this pathway downstream of GABAergic signals. Importantly, the authors show that activation of SMAD signaling with TGF-β or BMP4 potently decreased IL-13–induced goblet cell differentiation. So even during an inflammatory response, SMAD activation can block mucus cell metaplasia/hyperplasia. These studies highlight another possible target for pharmacologic blockade of goblet cell development in chronic airway diseases.

Now, with our extensive understanding of the pathways that control goblet cell development in chronic respiratory diseases, why is there no therapy directed at blocking goblet cell differentiation?

Very few clinical trials have focused on mucus as a therapeutic endpoint. This lack of investigation most likely stems from the difficulty of assessing changes in mucus production, as it is very labor intensive, requiring either airway biopsy, rigorous collections, and/or biochemical analyses of sputum components. Moreover, estimating airway obstruction from mucoid impaction has been difficult. However, newer computed tomography imaging methods show that assessment of mucus in the airways is possible and should be adopted in future clinical trials as a practical, noninvasive approach to measure changes in mucus (15).

Other limitations to therapies directed at goblet cell differentiation and death include the lack of specificity of the drivers of these processes for the airway epithelium; thus, systemic treatments may have multiple effects. This is true for EGFR tyrosine kinase inhibitors currently used for cancer treatment, as they exhibit drug class toxicity due to the presence of EGFR in other organs (16, 17). Similarly, $GABA_A$ receptor inhibitors have various toxicities in the nervous system that limit their systemic use, and Bcl2 inhibitors aimed at increasing goblet cell apoptosis also have serious systemic toxicities (18, 19). The data presented by Feldman and colleagues suggest that therapeutic targeting of SMADs by activation is a potential treatment for mucus hypersecretion. Yet, the authors also highlight the complex effects of using TGF-β family members in the lung, linking these cytokines to concerns about opportunistic infection, inflammation, and fibrosis. These drawbacks to site-specificity may be overcome in the future through local application of therapeutics by inhalation, or by the development of airway epithelium–specific modes of activation.

Inhaled therapies are a smart approach to targeting mucus in the airways, but barriers to achieving optimal effects remain. Mucus was the primary outcome in a clinical trial of an inhaled EGFR antagonist in subjects with chronic obstructive pulmonary disease (COPD) (17). In that study, airway epithelial cells in biopsies of BIBW2948-treated subjects showed reduced EGFR signaling, but there was no effect on mucin stores, nor was there any change in goblet cell size or number. Yet, when individual goblet cells were analyzed, there was a correlation between reduced EGFR activation and lower goblet cell mucin in the group that received the higher drug dose, suggesting that more effective inhibition of EGFR could decrease airway mucus in patients with COPD. Unfortunately, there was a high rate of adverse outcomes, including declines in forced expiratory volume in 1 second. In addition to drug-specific limitations, inhaled agents in mucus hypersecretory diseases must be able to traverse the mucus layer to engage the epithelial cells, and this poses another barrier to effective drug delivery and uniform distribution.

Targeting mucus as a therapeutic endpoint would seem obvious, but monoclonal antibodies directed against IL-13 or IL-4R-α were never tested for their ability to affect goblet cells or mucus production in many clinical investigations of asthma and COPD (20, 21). These therapies were found to be effective in reducing disease exacerbations in subjects with eosinophilic disease, suggesting that they predominantly serve to reduce inflammation rather than limit mucus, as mucus is produced independently of an inflammatory phenotype. Differentiating between effects on mucus versus inflammation becomes convoluted, as reducing inflammation leads to reduced mucus production. Investigations are still needed to assess the long-term impact of blockade of IL-13 on airway remodeling, including on the basal numbers of goblet cells and mucus glands, because these changes will affect lung function.

The studies presented by Feldman and colleagues enrich the depth of our knowledge about goblet cell differentiation, and this is crucial for future development of pharmacologic interventions in diseases involving mucus hypersecretion. Along with these advances, as we test new therapies in airway diseases, there should be a push for studies that assess outcomes that include mucus production. ■

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Clemente J. Britto, M.D.
Lauren Cohn, M.D.
Section of Pulmonary, Critical Care, and Sleep Medicine
Yale School of Medicine
New Haven, Connecticut

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