Understanding the Relevance of the Mouse Cigarette Smoke Model of COPD: Peering through the Smoke

Chronic obstructive pulmonary disease (COPD) is one of the most important threats to public health because of its prevalence, economic cost, and impact on death and disability. Although numbers are important, they do not begin to describe the personal toll felt by those with COPD who are dependent on inhalers, tethered to oxygen lines, isolated in their homes, and deprived of life's potential. Despite the importance of COPD, current therapies are woefully inadequate because they have only a modest effect on symptoms and exacerbations, and no effect on the long-term decline in lung function or death. The need for better therapies is palpable.

When testing a new idea, animal models are one of the first tools investigators turn to, because they have advantages over cell-based assays or direct human testing. For example, animal models allow mechanistic research to be carried out within the complexity of the whole organism, use of genetic methods to more precisely examine molecular mechanisms, and studies of drugs for safety and efficacy.

Cigarette smoke exposure has become one of the most utilized animal models of COPD. Long-term cigarette smoke exposure was shown to cause the morphologic and physiologic manifestations of emphysema first in guinea pigs (1) and later in mice (2, 3). Exposure of mice to cigarette smoke increases lung inflammation, protease activity, oxidant stress, and apoptosis, and in select strains results in the development of modest amounts of emphysema and mild degrees of airway and pulmonary vascular remodeling (4–6). On the other hand, cigarette smoke-exposed mice do not develop the excessive mucus production, mucus cell metaplasia, or periodic exacerbations/flares that characterize COPD in humans (4). This may be due, in part, to differences between mice and humans in lung development, airway branching, distribution or presence of respiratory bronchioles, basal cells, ciliated cells, goblet cells, club cells, or submucosal glands (4, 7). Despite these differences, the murine smoke exposure model is the most commonly used COPD model because experimental intervention in the regulation of molecular pathways is relatively easy, immunologic reagents are plentiful, and costs are low. But are these advantages enough for the murine smoke model to have such prominence in COPD research; that is, will it lead to better treatments?

In this issue of the *Journal*, Yun and colleagues (pp. 47–58) look for common genes and pathways that could explain differences in the susceptibility (or resistance) to develop COPD/emphysema for mice and humans (8). They exposed emphysema-susceptible and -resistant mouse strains to cigarette smoke for 6 mo, analyzed whole-lung gene expression, and compared these results to gene expression from the lungs of nonsmokers and former smokers, with or without COPD. The effect of smoke exposure on gene expression was tested in wild-type mice that were susceptible (C57BL/6) or resistant (NZW/LacJ) to the development of emphysema, as well as in susceptible ($Hhip^{+/-}$) or resistant ($FAM13A^{-/-}$) genetic mouse strains.

The results confirmed that cigarette smoke increases the expression of genes that regulate xenobiotic metabolism and the Nrf2 oxidative stress response. More of these genes were up-regulated in emphysema-resistant $FAM13A^{-/-}$ mice versus susceptible C57BL/6 or $Hhip^{+/-}$ mice, indicating that resistance to the development of emphysema may be related to a more robust xenobiotic and antioxidant response. The authors looked for candidate emphysema-resistance genes by examining the response of susceptible $Hhip^{+/-}$, moderately susceptible C57BL/6, and resistant $FAM13A^{-/-}$ mice to cigarette smoke exposure. This approach identified seven candidate resistance genes with expression levels that went up as the susceptibility to develop emphysema went down. In contrast, only baseline gene expression was different in emphysema-resistant NZW/LacJ mice versus susceptible C57BL/6 mice, suggesting that resistance may be affected by baseline gene expression.

The greatest strength of this study is the comparison of gene-expression profiles in human and mouse lungs as they relate to smoking and the development of COPD/emphysema. Their study found many overlapping genes that were associated with cigarette smoke, but far fewer that were associated with COPD. When specific mouse models were compared to human lungs, concordance of gene expression was better between humans with COPD and susceptible $Hhip^{+/-}$ mice than with resistant $FAM13A^{-/-}$ mice. Ultimately, only three human genes associated with COPD and emphysema overlapped with genes that were associated with emphysema in mice.

So how do we interpret these results? At first glance, the murine cigarette-smoke–exposure model appeared to have performed poorly. In view of the fact that cigarette smoke does not cause chronic bronchitis or substantial airway remodeling in mice, it seems prudent to consider using other models that may more faithfully mirror human disease, such as the guinea pig or ferret smoke-exposure models (4, 6, 9). Nonhuman primates also have clear relevance to humans (10), but come with distinct ethical and cost disadvantages that limit their general use for COPD research (11, 12).

The study also has limitations, including the fact that diseased human lungs were taken from patients with end-stage COPD. In humans, COPD develops slowly over decades as airways remodel and airspaces enlarge. Mechanisms that are active during this formative period may be very different from pathways that dominate in destroyed lungs. Gene expression was assessed in whole-lung homogenates using array technology, rather than on the single-cell level using the whole transcriptome. The authors did not confirm key results with PCR. They also did not control for the length of smoking cessation in mice or humans, which has a progressive effect on gene expression (13).

This study provides further evidence of what we already knew, namely, that mice do not completely reflect how COPD develops in humans, and should not be viewed in isolation. To develop effective

therapies that impact COPD patients in a meaningful way, we need to better understand if and where the pathobiology of mice is relevant to human disease, develop alternative animal models with powerful genetic and immunologic tools that more accurately reflect human disease, and learn how to identify early, prediagnostic stages of COPD in humans so that we can compare animal models with lungs that are still developing disease (14). Hopefully, these advances will give us the knowledge and tools to create therapies that improve the daily lives of millions with COPD. ■

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R. William Vandivier, M.D.
Division of Pulmonary Sciences and Critical Care Medicine
University of Colorado
Aurora, Colorado

Moumita Ghosh, Ph.D.
Division of Pulmonary, Critical Care and Sleep Medicine
National Jewish Health
Denver, Colorado

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