Crossing Kingdoms: Host–Microbial Endotyping and the Quest to Understand Treatable Traits in Chronic Obstructive Pulmonary Disease

The history of chronic obstructive pulmonary disease (COPD) is long. As eloquently reviewed by Thomas Petty (1), knowledge of its pathological components has evolved greatly the last two centuries. This evolution has accelerated in recent years with further delineation of COPD's heterogeneity (2–4). We now have more insight into the molecular basis for various clinical presentations and potential biomarkers (5–7). Ultimately, it is hoped that such advances lead to a better definition of endotypes and understanding of treatable traits, wherein we link specific mechanisms, biological markers of such, and, ideally, predictable response to treatments. An example of such is alpha-1 antitrypsin deficiency, a clear genetically linked endotype that can be treated with targeted augmentation therapy. Much remains to be worked out, but we are getting closer.

A COPD phenotype defined by eosinophilic or neutrophilic inflammation has been a focus of recent attention. These patterns have been linked to disease severity, outcomes including exacerbations, and response to treatment in particular inhaled corticosteroids (4, 7–9). However, the underlying factors driving these inflammatory patterns remain incompletely understood. Recent interest in the airway microbiome has grown out of preceding knowledge that sputum cultures from patients with COPD often yield potentially pathogenic bacteria that contribute to exacerbations and drive airway inflammation (10). Recent studies have described complex dynamics of the airway microbiota represented in sputum in the setting of exacerbations, a timing when this evaluation might be complicated by treatment confounders (11, 12). Exploration of how immune responses relate to airway microbiota patterns, particularly during clinical stability and over time, may yield new insights into the basis for different inflammatory phenotypes.

In this issue of the *Journal*, Wang and colleagues (pp. 1488–1502) pursue this, taking advantage of such data previously generated from three UK COPD cohorts (COPDMAP, AERIS, and BEAT-COPD) (13). Data from a total of 510 participants were available, including sputum bacterial profiles, cell counts, and measured immune mediators. They were able to pursue integrated microbiota analysis in two of the cohorts because sequencing was performed using the same procedures and platform (Illumina MiSeq) at a centralized laboratory, and the third cohort was used as validation, as the microbial data were generated differently. Sophisticated analysis approaches were employed to examine the primary question of whether neutrophilic and eosinophilic COPD associate with different airway microbiota profiles. Interestingly, neutrophilic COPD was associated with two main patterns of airway bacterial ecology defined by the predominance (or not) of *Haemophilus* members. *Haemophilus*-predominant neutrophilic COPD was relatively stable over time, irrespective of exacerbations, and associated with elevated sputum IL-1b and TNF-α. In contrast, the neutrophilic non–*Haemophilus*-predominant subgroup had a more balanced bacterial profile, wherein *Veillonella* and *Prevotella*, ubiquitous commensals of the oral cavity, were the most represented and associated with elevated IL-17A. This "balanced" subgroup displayed a tendency for greater shift during exacerbations, switching to either the *Haemophilus*-predominant neutrophilic or eosinophilic phenotype, which otherwise were mutually exclusive. Eosinophilic COPD displayed elevated type 2 inflammation markers and the airway microbiota characterized by greater representation of *Campylobacter*, *Granulicatella*, and *Gemellaceae*, among others. Several of these correlated with sputum eotaxin-3, IL-5, and TARC. Change-point analysis identified *Haemophilus* being associated with switches from eosinophilic to neutrophilic state during clinical stability. *Campylobacter*, *Gemellaceae*, *Capnocytophaga*, and *Granulicatella* were associated with switches to eosinophilic inflammation.

This is one of the largest studies to date examining relationships between the airway microbiome, immune markers, and the COPD inflammatory phenotype. The study also represents an important attempt at performing meta-analysis of airway microbiome data using information from three cohorts. This is no small feat given the challenges when attempting to harmonize different original workflows, from sample preparations to sequence data generation and provenance in analysis methods (14). In general, transparency of information remains an obstacle to integrating microbiome data from different studies, yet this is an important goal given the heterogeneity of such data and of the diseases under study. Here, the collaborative process was aided by similarities in cohort design and overall awareness of potential caveats. The results provide further insight into the potential role of airway microbiota in the COPD inflammatory phenotype, either as proximal drivers or microbial markers of such. That neutrophilic COPD associated with at least two different airway ecology patterns and dynamics indicates this phenotype does not always relate to airway dominance by pathogenic bacteria typically associated with COPD. The predominance of oral commensals in the neutrophilic-"balanced" subgroup is reminiscent of an earlier study in which lung microbiome enrichment with similar bacteria was associated with T-helper 17 cell related lung inflammation (15). In contrast, eosinophilic COPD was characterized by

Supported by National Institute of Allergy and Infectious Disease grant R01AI129958 (Y.J.H.) and National Cancer Institute grant R37CA244775 (L.N.S.).

Originally Published in Press as DOI: 10.1164/rccm.202101-0169ED on March 2, 2021

enrichment in bacteria not usually considered pathogenic in themselves. Interestingly, the bacterial ecology in this group was similar to that observed in sputum from allergic adults with or without mild asthma (16) who had a similar range of blood, but not sputum, eosinophils as the subjects with COPD here. This raises the question of whether and to what extent underlying allergy may contribute to eosinophilic COPD given the overlapping biological features.

Wang and colleagues analyzed data from an impressively large set of samples. Notwithstanding the importance of this effort, several challenges and limitations are noted. The study was a retrospective analysis of previously generated data, and time-series analysis was feasible in only a small subset of the total combined cohort. More importantly, the subjects largely represented those with more advanced symptomatic COPD, the majority of whom are on inhaled corticosteroids, drugs that can affect the assessment of inflammatory markers and microbes. Thus, the extent to which the findings reflect molding of the airway environment over time versus biological interactions that may impact earlier stages of COPD pathogenesis remains unclear. Sputum is more practical to collect in large studies and during exacerbations but inherently includes a mixture of both upper and lower airway components. Thus, the microbial–immune relationships observed here may not be reflected in lung samples collected by bronchoscopy or be additionally impacted by the method of sputum collection. A recent study of daily sputum collected from patients with cystic fibrosis during clinical stability found day-to-day variation in airway bacterial community structure (17). This has implications for deriving and incorporating microbiome biomarkers into schema for disease phenotyping or prognostication.

Nonetheless, the current study by Wang and colleagues is an important contribution to the field. It demonstrates how to thoughtfully approach integrated analysis of microbiome data, whether melding such across different studies or combining it with other types of high-dimensional data, efforts increasingly being pursued across studies of respiratory disease. These integrative efforts might seem right now as an analytical tour-de-force but are necessary steps to build a microbial–host biomarker approach that earmarks specific endotypes in complex diseases such as COPD. We need such efforts to better understand specific treatable traits in COPD, improve personalization of treatment, and help move away from trial-and-error therapy with a limited number of drug combinations. ■

Author disclosures are available with the text of this article at www.atsjournals.org.

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