function and may represent an opportunity for intervention that restores health or abrogates progression to disease.

To move toward these aims, we convened a working group in July 2012 and a workshop in September 2013 to discuss the state of the art, identify key issues and knowledge gaps, and develop a strategy for promotion of lung health and prevention of lung diseases. Experts in the areas of lung health, asthma, bronchopulmonary dysplasia, chronic obstructive pulmonary disease, cystic fibrosis, interstitial lung disease, and pulmonary hypertension discussed this currently underemphasized aspect of lung research. While it was apparent that all disease areas could benefit from multidisciplinary research to develop markers of health and disease, identify targets for intervention, and test preventive concepts, researchers in some disease areas are already beginning to consider primary prevention interventions. Although disease prevention research was more mature in some areas than others, the principles of how to approach this major new effort are likely to be similar across the range of lung diseases. The conclusions and recommendations from the seven topics of primary prevention research are presented in the April supplement to the *Annals of the American Thoracic Society*, which will be published online on April 15, and will serve as the basis for the development of a staged strategy for primary prevention research by the NHLBI.

Major themes in this effort will be the definitions of lung health, preclinical, and disease states, which vary both on a biological continuum and over time. The concept of lung health, while consistent with the Barker hypothesis for the fetal origins of adult disease, must also consider biologic, behavioral, environmental, and socioeconomic influences on health and disease risk during the life of an individual or population, and even across generations. Emerging evidence indicates that environmental, behavioral, and socioeconomic influences impact respiratory and immune biology in a sustained manner that can accumulate and positively or negatively affect lung health outcomes for individuals and populations. Lung health is a complex concept that must expand beyond currently defined physiologic tests, and should be measurable at both individual and population levels. Temporal effects may be more complex than simply characterizing lung development and growth from gestation into adolescence and young adulthood; the life course of lung health likely includes critical periods when an exposure can have sustained effects on function and risk can accumulate over time or even across generations. Better understanding and definition of lung health, during the entire lifespan, promises to identify opportunities to intervene at pivotal points to increase and/or preserve lung health. Such interventions may be at the population or societal level as well as the level of the individual. It may be possible to both promote development of healthier lungs and slow decline of aging lungs. Even small improvements of average respiratory health (as defined by multiple measures) of a population will have a significant public health impact immediately and could be compounded over future generations.

A related important theme is the critical need for a better definition of "disease." Disease is typically tied to clinical variables and a doctor diagnosis, but the pathobiology that leads to disease is dynamic and on a continuum from health to detectable abnormalities (e.g., imaging and serum markers) to disease. As we gain a more integrated understanding of lung health–incorporating molecular, cellular, organ, and system information, it will become possible to identify critical risk factors and biologic events that represent a disruption in health and homeostasis. Comprehensive signatures of risk and perturbed healthy processes should be developed that define preclinical disease states as well as the points of disease origin, because these likely will illuminate modifiable targets for intervention in both general and disease-specific primary prevention strategies. Understanding susceptibility and inception of disease in susceptible versus "protected" individuals may also be revealing of beneficial approaches for promotion and optimization of respiratory health and primary prevention of lung disease.

We challenge ourselves and the pulmonary community to join together and create a new paradigm in which optimal, sustained respiratory health and primary prevention of lung diseases are viewed as feasible and practical goals at both the individual and population levels. Multidisciplinary approaches and teams will be needed for these investigations, and great creativity will be required for research that produces effective preventive strategies. NHLBI will continue to work, in partnership with our pulmonary community, to identify gaps, opportunities, and the best approaches in preventive research. We believe that primary prevention of chronic lung diseases is an achievable and obligatory goal for our nation. ■

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Cystic Fibrosis Transmembrane Conductance Regulator and *Pseudomonas*

Cystic fibrosis (CF) lung disease is characterized by chronic airway infection and a heightened immune response leading to airway damage and bronchiectasis (1). *Pseudomonas aeruginosa* is the species of bacteria most commonly associated with CF, with almost 80% of adults chronically infected (2). Chronic *P. aeruginosa* infection is associated with more rapid decline in lung function and decreased survival (3, 4). Inhaled antipseudomonal antibiotics are effective in eradicating early *P. aeruginosa* infection and appear

to delay the onset of chronic *P. aeruginosa* infection (5). Current treatment guidelines focus on early detection and attempted eradication of *P. aeruginosa* prior to establishment of chronic infection (6). Once chronic infection develops, eradication is rarely achieved, and the goals of inhaled antibiotic therapy change to bacterial load suppression rather than eradication. Despite chronic infection, resistance to eradication by antibiotics, and high airway bacterial loads, invasive *P. aeruginosa* infection is rare in CF (7).

The primary defect in CF is absence or diminished production of the CF transmembrane conductance regulator (CFTR) protein due to mutations in the CFTR gene (1). CFTR protein functions as a cell membrane channel responsible for chloride and other ion transport across epithelial cell linings in the lungs, gastrointestinal tract, and pancreas. Lack of CFTR in the lung is thought to lead to a dehydrated airway surface liquid layer, mucus abnormalities, and altered immune response. Efforts to treat *P. aeruginosa* infection have been hampered by a lack of understanding of how defects in CFTR protein lead to chronic *P. aeruginosa* infection. Several mechanisms have been proposed. Arguably, the most accepted hypothesis is that dehydration of the airway surface liquid due to sodium hyperabsorption leads to impaired mucociliary clearance, bacterial stasis, and poor clearance of bacteria; the propensity for *P. aeruginosa* infection, however, is not well explained by this mechanism (8). Increased adherence of *P. aeruginosa* to CFTR-deficient airway epithelial cells has been proposed, but results of experimental studies are mixed, and most *P. aeruginosa* found *in vivo* is not attached to the epithelium (9). CFTR dysfunction has also been implicated in the exaggerated proinflammatory, neutrophil-dominated, immune response seen in patients with CF, but why this immune response is ineffective at eradication *P. aeruginosa* is incompletely understood (10). Biofilm formation by bacterial species such as *P. aeruginosa* has been proposed as a mechanism of chronic infection and poor susceptibility to antibiotics. However, *P. aeruginosa* isolates from patients with CF with chronic infection are variable in their ability to form biofilms. Recently aggregates of *P. aeruginosa* similar in characteristics to biofilms but nonadherent have been proposed as a mechanism of chronic infection in CF (11).

Clinical observational data clearly supports a link between CFTR dysfunction and chronic *P. aeruginosa* infection. Chronic *P. aeruginosa* is more common in those with absent CFTR function (two severe CFTR mutations) compared with those with residual CFTR function (one or two mild CFTR mutations). Even in early disease, risk of *P. aeruginosa* acquisition is increased in those with absent CFTR function (12). In addition, individuals with non-CF bronchiectasis develop chronic *P. aeruginosa* infection at lower rates compared with those with CF-related bronchiectasis (13). CFTR dysfunction thus appears to favor chronic *P. aeruginosa* infection that is difficult to eradicate. Understanding the link between CFTR and *P. aeruginosa* may lead to novel treatment approaches.

In this issue of the *Journal*, Staudinger and colleagues (pp. 812–824) address several key questions regarding the link between CFTR and chronic *P. aeruginosa* infection (14). They first investigated how airway conditions associated with CFTR dysfunction, increased mucus density, and altered biochemical properties of sputum might lead to the phenotypic eradication-resistant, antibiotic-tolerant, and noninvasive chronic *P. aeruginosa* infection in CF. They specifically focused on the growth of bacteria in aggregates rather than biofilms, and explored airway and bacterial features that would promote aggregate formation. Using models of low- and high-density agar gels, plus the addition of soluble components of CF sputum, they found that both high-density gels and soluble sputum components promoted *P. aeruginosa* growth in aggregates. Hypothesizing that neutrophil elastase (NE) within the sputum may drive aggregate formation, they performed a series of experiments to disrupt NE activity within sputum, restore activity using purified NE, and then disrupt NE again with elastase inhibitors. Aggregate formation occurred in the presence of NE but was prevented when NE activity was diminished. Using genetically immotile *P. aeruginosa* and motility studies of bacteria exposed to NE, they showed that decreased bacterial motility may be the common mechanism by which increased density mucus and NE lead to aggregate formation.

Staudinger and colleagues also showed that *P. aeruginosa* grown in aggregates had increased resistance to killing by NE and antibiotic tolerance compared with dispersed bacteria. Tobramycin appeared to provide a competitive advantage to nonmotile *P. aeruginosa*. These studies suggest that aggregate formation may increase antibiotic tolerance even in genetically susceptible bacteria and that chronic use of antibiotics may push infections toward this more tolerant phenotype. Importantly, genetic mutations preventing biofilm formation, found in many late CF *P. aeruginosa* isolates, did not prevent aggregates from developing. To explore whether aggregate formation could reduce invasiveness, they studied the effect of aggregates on epithelial cells and in an *in vivo* wound model and found attenuated immune response and decreased mortality in the presence of aggregates. Thus, conditions in the CF airway due to CFTR dysfunction and resultant treatment, including dense mucus, high levels of NE, and exposure to chronic antibiotics, appear to favor nonmotile *P. aeruginosa*, promoting aggregate formation. These aggregates may at least partially explain the eradication-resistant, antibiotic-tolerant, and noninvasive infection seen in CF.

Despite advances in early eradication approaches and chronic suppressive therapy with inhaled antibiotics, *P. aeruginosa* infection remains a significant problem in CF and contributes to much of the morbidity and mortality. The work by Staudinger and colleagues provides important insight into how the basic defect in CF, CFTR dysfunction, may foster an environment ideal for chronic *P. aeruginosa* infection. Yet, we are still left with the question of why the CF airway is susceptible to *P. aeruginosa* in particular. One limitation of the study is that it focuses exclusively on *P. aeruginosa*, whereas many other bacteria, including *Staphylococcus aureus*, other gram-negative bacteria, nontuberculous mycobacterium, and, more recently, obligate anaerobes, are known or suspected to contribute to CF lung disease (15, 16). Infection is frequently polymicrobial; thus, how polymicrobial infections impact aggregate formation is an area that needs further study. Another avenue of research is whether restoration of CFTR function can reverse the pressure on *P. aeruginosa* to form aggregates once chronic infection has developed. As demonstrated by Staudinger and colleagues, the mechanisms linking CFTR dysfunction and chronic *P. aeruginosa* infection are complex and multifaceted. Perhaps our best hope for

true eradication of *P. aeruginosa* is through improved CFTR function, a possibility now with the development of CFTR modulators. ■

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The Burden of Disease and the Need for a Simple Staging System in Idiopathic Pulmonary Fibrosis

Recently a debate has emerged regarding the optimal endpoint for phase 3 clinical trials in idiopathic pulmonary fibrosis (IPF). IPF clinical trials, including the pirfenidone and nintedanib (BIBF1120) trials, have used FVC as a surrogate endpoint for demonstrating drug efficacy, with a goal of achieving regulatory approval (1). Raghu and colleagues have published a consensus view from a summit of North American key opinion leaders suggesting that all-cause mortality or a composite of all-cause mortality and all-cause nonelective hospitalization were the most clinically meaningful endpoints for phase 3 clinical trials in IPF (1). This view on mortality as a clinical trial endpoint has been opposed by another international panel (n = 52) of experts on IPF, who state that the use of mortality as an endpoint in IPF trials is impractical and sets a standard not required for drug registration in other respiratory diseases, including lung cancer, pulmonary arterial hypertension, cystic fibrosis, and chronic obstructive pulmonary disease (COPD) (2). In this issue of the *Journal*, King and colleagues (pp. 825–831) address the question of the use of mortality as an endpoint, by analyzing the mortality data from the placebo arm data of the INSPIRE and CAPACITY trials in IPF (3). The authors calculate that the estimated number of subjects necessary to perform a trial enrolled over 3 years, to detect a 25% reduction in mortality as the primary endpoint, with 90% power and up to 5-year follow-up, in a theoretical, randomized, double-blind, placebo-controlled drug trial in patients with IPF, would be 2,582 patients. This is not financially viable and obviously impractical.

King and colleagues begin their article by highlighting that IPF is a fatal condition, yet conclude that mortality is not a valid endpoint (3). This initially appears to be a contradiction in terms. The inconsistency is related to the simple concept that survival is a function of the burden of disease. The population studied in the placebo arm of the current study represents mild to moderate physiological impairment, which indicates a limited burden of disease. Had the placebo arms of an alternative phase 3 clinical trial in IPF been used, for example, the STEP-IPF study, the power calculations would likely be significantly different (4). The mortality rate at 28 weeks in this study was 13% (11/91), as compared with the mortality rate of 13.7% at 2 years in the cohort