Cystic Fibrosis Therapy: A Community Ecology Perspective

Douglas Conrad¹, Matthew Haynes², Peter Salamon³, Paul B. Rainey⁴,⁵, Merry Youle⁶ and Forest Rohwer⁷

¹University of California San Diego, Department of Medicine
²DOE Joint Genome Institute, Walnut Creek, CA
³San Diego State University, Department of Mathematics
⁴Massey University, New Zealand Institute for Advanced Study & Allan Wilson Centre for Molecular Ecology & Evolution
⁵Max Planck Institute for Evolutionary Biology
⁶Rainbow Rock, Ocean View, HI, USA
⁷San Diego State University, Department of Biology

Corresponding Author: Douglas J. Conrad
Division of Pulmonary and Critical Care Medicine
University of California, San Diego
San Diego VA Healthcare System
3350 La Jolla Village Drive, 111J
San Diego CA 92122
dconrad@ucsd.edu

Acknowledgements:
The authors would like to acknowledge support of the Cystic Fibrosis Research, Inc. (CFRI) grant no. 55676 and NIH RO1 56586A.
Abstract

Current therapy for cystic fibrosis (CF) focuses on minimizing the microbial community and the host’s immune response through aggressive use of airway clearance techniques, broad-spectrum antibiotics, and treatments that break down the pervasive endobronchial biofilm. Antibiotic selection is typically based on the susceptibility of individual microbial strains to specific antibiotics \textit{in vitro}. Often this approach cannot accurately predict medical outcomes due to factors both technical and biological. Recent culture-independent assessments of the airway microbial and viral communities demonstrate that the CF airway infection is considerably more complex and dynamic than previously appreciated. Understanding the ecological and evolutionary pressures that shape these communities is critically important for the optimal use of existing therapies (both the choice of therapy and timing of administration) and the development of newer strategies. The Climax-Attack Model (CAM) presented here, grounded in basic ecological principles, postulates the existence of two major functional communities. The Attack community consists of transient viral and microbial populations that induce strong innate immune responses. The resultant intense immune response creates microenvironments that facilitate establishment of a Climax community that is slower growing and is inherently resistant to antibiotic therapy. Newer methodologies, including sequence-based metagenomic analysis, can track not only the taxonomic composition but also the metabolic capabilities of these changing viral and microbial communities over time. Collecting this information for CF airways will enable mathematical modeling of microbial community dynamics during disease progression. The resultant understanding of the airway communities and their effects on lung physiology will facilitate optimization of CF therapies.

\textbf{Key Words:} Cystic fibrosis, airway ecology, metagenomics
Introduction

Our view of cystic fibrosis (CF) airway infection is shifting from one based on the analysis of single microbial strains under laboratory conditions to one grounded in an ecological, systems-based assessment of entire communities interacting in situ with the host immune response. Laboratory studies of single pathogens provide valuable information about their potential roles in CF; however, colonization of the CF airway is not achieved by a single strain or species acting in isolation. The CF airway is a complex ecosystem hosting a heterogeneous community including bacteria, viruses, and fungi (1-3). Immigration (colonization) events, the order of arrival, and the ensuing interactions between microbial strains further complicate the work of the physician treating a CF patient. Future breakthroughs in the control of infection in the CF lung will most likely come as a result of our increased understanding of the ecological and evolutionary processes shaping these microbial communities (4, 5).

Here we advocate for an ecological perspective that recognizes the chronic CF airway infection as a polymicrobial community and that informs new treatments consistent with established ecological principles. In this light, CF infection is no longer viewed as the invasion of a single pathogen, such as Pseudomonas aeruginosa, that could be countered by the administration of antibiotics targeting that organism. Instead, disease is seen as the result of dynamic changes in the airway microbial populations, with concomitant alteration of the community physiology. Such a perspective focuses our attention on the factors that regulate microbial community stability and function, the nature of interactions between the bacterial and phage populations, and the influences that control microbial populations. The well-studied responses of the host's innate and adaptive immune systems are now seen to be acting within this dynamic ecological theatre and, in turn, to be eliciting evolutionary responses from the microbial community—all of which impacts the health of the host.
This ecological perspective has important therapeutic implications, offering the prospect of novel approaches to treatment. For example, instead of antibiotics directly targeting a specific pathogen, control of chronic airway infection might be achieved by disturbing some factor within the lung that is essential for growth and persistence of the pathogenic organisms, such as the presence of another community member, a nutrient, or other environmental attribute. Alternatively, the patient's immune responses might be manipulated so as to favor a shift of the composition and function of the microbial community toward a less pathogenic state. Successful implementation requires the ability to predict and manipulate the outcomes of potential clinical treatments. As a step toward that goal, we propose a model, grounded in ecosystem concepts (6, 7), of the microbial communities within the lungs of CF patients. Such a model can optimize the use of current therapies as well as indicate potential novel therapeutic approaches. It also provides a framework for interpreting the results of past research and clinical experience, and can set the direction for productive future investigations.

The Microbiota of the CF Lung Ecosystem: During CF infection, the lungs house a complex ecosystem consisting of dynamic communities of bacteria, fungi, and viruses, many—possibly most—of which are yet to be characterized. The accumulation of mucus in the bronchial tree creates an environment conducive to colonization by various opportunistic pathogens. Initially the community is dominated by aerobes, succeeded later by facultative and strictly anaerobic microbial populations (8). Thus, in younger CF patients, Staphylococcus aureus, Haemophilus influenzae, and P. aeruginosa are common, whereas in older patients, P. aeruginosa dominates the ecosystem, often accompanied by other pathogens such as S. aureus, Burkholderia cepacia complex, Stenotrophomonas maltophilia, and Achromobacter spp. Recently, large numbers of anaerobic bacteria have been identified in the sputum of CF patients, although their clinical significance remains uncertain (9). Additionally, non-tuberculous Mycobacterium spp. have been isolated in CF patients. Their frequency, ranging from <10% up to >20% in different regions of the USA, may reflect the varying environmental prevalence of these organisms (10-13). Among the fungi present,
Aspergillus spp. and Candida spp. are most frequently observed, but more drug-resistant species, such as Scedosporium azoospermia, are becoming common (14). The viruses include both those that potentially infect humans as well as bacteriophage (phage) that prey upon the bacterial community (15).

In the past five years, culture-independent assessments of the microbial and viral communities in CF airway secretions (sputum and bronchial alveolar lavage fluid) have revealed a more varied community than had been evident using standard microbial culture techniques. Based on the 16S rRNA sequences present, this approach has shown not only that the microbial communities are more diverse than previously appreciated, but that this diversity may peak during adolescence, prior to significant structural remodeling of the lung (16, 17). Recent metagenomic studies characterizing the microbial and viral communities within lung tissue dissected from distinct lung regions added the spatial dimension, reporting that different lobes housed distinctly different communities—thus further complicating clinical treatment (15, 18).

Control of Microbial Populations: Effective, ecologically-based CF treatment could minimize lung damage and loss of function by reducing or altering the microbial communities present in the lung. Current clinical therapies such as airway clearance therapy and the administration of antibiotics directly—but often indiscriminately—decrease the microbial populations. The human immune system also exerts powerful top-down control of microbes in the lung, but its actions have detrimental long-term repercussions. Although it protects the lungs in the short term by killing microbes, during chronic CF airway infections, the immune response maintains ongoing inflammation. This, in turn, leads to airway remodeling and the creation of a spatial structure with ensuing effects on microbial diversity and community stability that are likely to make microbial populations more difficult to eradicate (19).
Lung bacterial populations are also subject to top-down control by phage. Phage are the most abundant entities in all characterized ecosystems, with typically ~10 phages for every bacterial cell. Since most phage quickly kill their bacterial hosts, they are responsible for significant bacterial mortality. Unlike other agents of top-down control, phage are selective predators, each type infecting only related strains/species of bacteria. Thus, their top-down control also impacts the relative abundances of bacterial populations (20). They have been isolated from various sites within the human body, including airway secretions (20-25) and lung tissue (15) of CF patients. Significantly, phage were found to be absent from the late stage biofilms in CF lungs, suggesting that biofilm formation may help bacteria escape this top-down control. Further characterization of the phage community and its impact on the bacteria present in the CF lung can be expected to provide new insights into top-down control of the infection, including the potential use of phage for novel therapeutic options (26).

Other controls, termed **bottom-up controls**, limit growth and reproduction of microbial populations by limiting or partitioning essential resources. The CF airway provides a wide range of nutrients including amino acids, phospholipids, DNA, and a range of sugars (27), and any of these resources may fluctuate across space and time. This chemical complexity, along with spatial heterogeneity, affords ecological opportunity for bottom-up controls. It is likely that such resident controls are critical to the emergence and maintenance of microbial diversity in the CF lung (19, 28, 29). Potential therapeutic use of bottom-up controls might be in the form of the low concentration of a limiting micronutrient or the inhibition of a critical nutrient pathway in a specific environment. For example, inhibition of nitric oxide synthase in the CF airway may create a shortage of nitrite and nitrate, both of which are critical for the growth of denitrifying microbial populations (30).

**Landscape Ecology**: **Landscape ecology** considers the impacts of a heterogeneous environment on the organisms that reside therein. In the CF lung, this environmental heterogeneity is evident in the
patchy distribution of resources, the spatial structuring of the lung over distances both small and large, and regional differences in the microbial communities (18). A heterogeneous environment affords ecological opportunity; provided that resources are limited, ecologically diverse populations will evolve (30, 31). A heterogeneous environment also supports the maintenance of this diversity (19, 32). In contrast, a homogeneous environment limits both the opportunity for diversification and the level of diversity that can be maintained.

Interactions among the component organisms are central to the functioning of any microbial community (33-36). For example, excreted metabolites provide opportunities for both cooperative (syntrophic) and antagonistic (allelopathic) interactions. Small signaling molecules, such as the homoserine lactones or 4-hydroxy-2-heptylquinoline-N-oxide, allow populations to coordinate activities in a variety of antagonistic or mutually beneficial ways (37, 38). In the context of the CF lung, the full range of interactions is likely. The extent and nature of these interactions will reflect the degree of environmental heterogeneity as well as the level of connectivity that allows for the movement of organisms or resources from place to place.

How heterogeneous and independent are the macro-environments within the CF lung? Although each lobe constitutes a spatially distinct region, and the bronchioles are clearly spatially and physiologically separate from alveoli, the question remains to what extent microbes experience these as distinct niches. In other words, does the lung provide a well-mixed environment with resources and organisms dispersed uniformly throughout? Or is it a cluster of isolated niches? Understanding the extent of connectivity between populations within a single lung will influence how we think about the emergence of antibiotic resistant strains, the mucoid phenotype, and mutator strains within lung populations. Of these three, the evolution of antibiotic resistance is currently the most pressing issue. If connectivity between lobes is high, then physicians would be right to be concerned about the detection of an antibiotic resistant bacterial strain that could rapidly spread throughout the lung, supplanting susceptible types when under selection pressure from antibiotic treatment. If, instead,
the lung is spatially structured with little connectivity between niches, then an antibiotic resistant
type evolving, for example, in the right lower lobe would be unlikely to have the opportunity to
colonize the right upper lobe. Because phage in the CF lung harbor a reservoir of antibiotic
resistance genes (18), the phage-mediated spread of antibiotic resistance from lobe to lobe may also
be important.

**CF Disease States as Alternate Stable States:** For any ecosystem, there are many combinations of
biological and environmental conditions that can yield the same recognizable community, referred to
as a stable state (or an attractor). When a resilient ecosystem is perturbed, it recovers and returns to
its original "stable" state. However, a stronger perturbation may shift an ecosystem into an
alternative—also stable—state, that maintains a different community of organisms. Restoration
ecology seeks to identify the perturbations that cause ecosystems to shift from one state to another
and to then use appropriate perturbations to restore ecosystems to the more desirable state.

This same restoration approach can be applied to the CF lung. When a CF airway infection is
viewed as an ecosystem, one sees, in the simplest configuration, two alternative states: a quiescent,
clinically-stable state and an exacerbated state. The quiescent state resembles an ecosystem that is
dominated by stable populations that are well adapted to their particular biological niche.
Perturbations in the local environment that alter top-down or bottom-up population controls can alter
this stable community, possibly shifting it to an alternative state.

**The Climax-Attack Model of the CF Lung Ecosystem**

**Why Model?** The ecological principles discussed above suggest numerous potential
therapeutic strategies that might be efficacious in shifting the CF community to a less pathogenic
state. However, each tactic needs to be evaluated in advance to determine its optimal mode of
application and to predict possible adverse effects. Mathematical modeling of the interactions
between the CF microbial community and environmental factors can be used to test potential therapies. For instance, modeling may suggest that a particular therapy might be more effective in reducing overall microbial abundance when applied at a particular stage in the disease cycle, or it may identify therapies that can shift the CF airway ecosystem from a sick, exacerbated state to a healthier quiescent state. In this manner, the coupling of ecological modeling with methods from microbial ecology can lead us to better control of CF airway infections.

The Climax and Attack Communities:

Based on current ecological models and knowledge of the human airway microbial community, we posit the existence of two broad functional classes of CF microbial communities: a virulent, transient Attack community that is associated with pulmonary exacerbations and a Climax community that dominate during relatively stable periods (Figure 1). Our proposed ecological Climax-Attack Model (CAM) is based on the abundant, persistent “keystone” populations that characterize the microbial communities present in the quiescent and the exacerbated states (3, 39-42). Both Attack and Climax communities can coexist in a given patient; sequential Attack communities dominate the airway microbiota prior to significant airway remodeling, whereas the Climax communities prevail in heavily remodeled airways.

In the Climax-Attack community Model, a taxonomic unit such as P aeruginosa, as identified in the microbiology lab or by culture independent means i.e. 16s rRNA sequencing, can belong to both communities. For this reason, our model focuses not only on taxonomic characterizations but importantly on functional differences. These two communities are not the same as the statistically-defined “core” and “satellite” communities previously proposed (3, 43), but rather are defined by their metabolic capabilities, as determined by metagenomic, metatranscriptomic and functional microbiological studies. By sampling an Attack or Climax community and then sequencing the DNA from the microbes present, one generates a metagenome,
i.e., a set of gene sequences for that community, some of which can be assigned a known function. On this basis, it is appropriate to define an Attack metagenome as a microbial metagenome that is enriched for the virulence genes that elicit intense innate immune responses and subsequently initiate airway remodeling. Similarly, a Climax metagenome is enriched for genes that promote chronic colonization of the CF airways, elicit the antibody-dependent, adaptive immune response, and decrease antibiotic susceptibility. Metagenomic analysis is particularly advantageous here, as it does not limit investigation to identified cultured microbes. Instead it enables characterization of the metabolic capabilities of the entire microbial community within an individual patient—a community that is likely to differ in detail over time and between patients. A critical task facing CF-related metagenomic studies is to characterize the Attack and Climax metagenomes, to identify the changes that take place over time, and to correlate these changes with their clinical context, including therapy administered.

Recent studies demonstrate that the sputum during many pulmonary exacerbations is dominated by bacterial taxa present in the airways are similar to those in a clinically stable state (42). The Climax-Attack Model is consistent with these findings because the changes in the communities are functional changes and therefore are not necessarily identified by standard microbiological assessments or 16S rRNA sequence taxonomic assessments. For example, acute pulmonary exacerbations can result from aggressively growing revertants derived from slowly-growing, endogenous microbial populations or a change in bacterial gene expression eliciting stronger immune responses (44, 45) (Floyd Romesberg, personal communication). In either instance, the signs and symptoms of a pulmonary exacerbation could result without major changes in the dominating taxa during a more stable period. In other cases, the Attack communities associated with exacerbations are predicted to be dynamic and are frequently acquired through environmental exposure or aspiration, and include populations such as Streptococcus spp., Staphylococcus spp., non-mucoid Pseudomonas spp. and eukaryotic viruses (e.g., influenza, rhinoviruses). These
environmentally acquired strains are more obvious in early disease before established lung remodeling but certainly are known to be associated with succession events in patients with more advanced airway remodeling. In all cases, the Attack community may express a significant complement of wild type virulence factors, elicits strong responses from the innate immune system, and produces sudden illnesses and abrupt decline in lung health. The resultant clinically observed physiological changes are generally limited in time and space, and can be reversed by successful therapy. These changes result from both the immune response and the growth of biofilm in the airways, and include dyspnea, sputum production, cough, and systemic signs of inflammation. Remodeling is limited by the transient nature of the community and by successful exacerbation therapies. These dynamic Attack communities dominate the conducting airways of early stage CF patients prior to significant airway remodeling. However, their fast growth and immunogenic nature enable them to also initiate and propagate permanent airway remodeling, thereby creating the microenvironments that allow establishment of the chronic Climax community (46).

In contrast, the model predicts that Climax communities are composed of persistent, slow-growing microbial and fungal "keystone" populations whose metabolisms are well adapted to the CF airway microenvironment (31, 39, 44, 47). The organisms present include commonly cultured microbial populations such as mucoid *Pseudomonas* spp. (46), chronic *Staphylococcus* spp., *S. maltophilia*, *Achromobacter* spp., and other strict anaerobes. Island biogeography theory (48), supported by recent experiments (49), shows that community membership, function, and evolution depend also on the immigration history, including the sequence and timing of arrival of each organism.

The Climax community thrives in remodeled airways, ensconced within biofilms of their own making that generally provide protection from the patient's immune response. While this community does not cause sudden, transient drops in lung function, together with the Attack community it is responsible for the steady progressive loss of lung function over time. In most cases,
the pace of airway remodeling by the Climax community drives the long-term prognosis due to its prolonged stimulation of innate and adaptive immune responses, particularly evident when dominated by mucoid *P. aeruginosa* or *B cepacia* complex. For example, the destructive antibody response to the *Pseudomonas*-produced alginate is a major determinant of lung function in CF patients (44). The Climax community is generally resistant to standard antibiotic therapy and is best controlled by airway clearance therapies (e.g., inhaled hypertonic saline, inhaled recombinant human DNase) and, to a certain extent, by very high concentrations of inhaled antibiotics. Its rapid expansion is facilitated by the frequent exacerbations caused by Attack communities, by non-compliance with chronic therapies, or conceivably by the loss of other top-down controls, e.g., reduced lytic phage populations, impaired immune system function. Although Attack communities are usually associated with pulmonary exacerbations, a slow unimpeded expansion of the Climax community because of medical non-adherence is also associated with pulmonary exacerbation signs and symptoms.

**CAM and Clinical Therapies:** The Climax-Attack Model can inform the development of effective clinical protocols for different disease stages. For example, early in a patient’s life, the Climax community is small or non-existent; the microbial, viral, and fungal populations are primarily those characteristic of Attack communities. It is important to eradicate these early infections to minimize their impacts and delay the development of the Climax community. At some point, though, the cumulative damage allows the establishment of a Climax community. Based on the Climax-Attack Model, one can predict that treatments will have different effects on the two communities. For example, the Attack community would be expected to be more susceptible to antibiotics. Thus, broad-spectrum antibiotics used to kill susceptible members of the Attack community will select and favor the growth of the non-susceptible microbes present.
Based on the Climax-Attack Model, we would expect there to be varied possible etiologies for CF pulmonary exacerbations. This model enables us to relate exacerbations to various factors such as the patient’s clinical history, physiology, and airway community function as revealed by metagenomic analysis. For example, expansion of the Climax community would be predicted to occur during periods of therapeutic noncompliance or inefficacious therapy in patients with significant pre-existing airway remodeling. A sharp decline in lung function due to acquisition of virulent environmental strains would be predicted to have a metagenomic functional profile that would be distinct from that during exacerbations due to secondary viral infections, inhalation of toxic substances (e.g., smoke, dust), etc.

**Mathematical Models of Microbial Communities in the CF Lung:** The ecological Climax-Attack Model (CAM) provides a framework for modeling specific aspects of the CF airway disease. Such models assist in the interpretation and synthesis of prior observations and form a coherent basis for the development and evaluation of new therapies. Any useful model must address both the cyclical nature of the disease progression and the dynamic spatial heterogeneity of disease states throughout the lung. We discuss three approaches to modeling CF lung disease: *statistical* models that extract significant correlations from the CF Foundation Patient Registry; *physiological* models that correlate the degree of airway remodeling with decreasing lung function and increasing age; and *community dynamics* (ecological) models that express the spatially and temporally complex interactions between the host, the bacteria, and the phage.

**The Statistical Model:** The CF Foundation Patient Registry and individual case data are excellent sources for longitudinal data that include repeated observations on the same individuals. For example, registry data has been used to examine the effects of age and gender as predictors of future FEV1 (50). As a first step in developing this model, data mining and exploratory data analysis of registry data would be used to classify cases based on the severity of the disease and to identify
the key factors driving CF lung disease within each class. As one example, this approach can identify the variables that best predict changes in FEV1 over time. A similar study of pediatric CF patients using European patient data examined many variables; however, derived variables such as regression coefficients were not considered. The process of building a linear mixed model such as this for longitudinal data is an iterative process that requires a series of model fitting steps and repeated searching for the most significant correlations among the data (51). This strategy balances statistical and biomedical considerations with a goal of identifying disease classes and the most physiologically significant factors for each class.

**The Physiological Model:** A physiological model tracks the persistent effects of the Attack community (scarring due to the host inflammatory response) and the growth and spatial distribution of the Climax community by modeling biofilm expansion, increased mucus plugging, and altered airway function over time. The model is based on the fractal structure of the lung (52-54). It assumes that the rate of mucus accretion in an infected bronchiole is a constant characteristic of the local Attack community. When the accumulating mucus fills the bronchiole, the passage becomes blocked, thus making all distal lung volume inaccessible and thereby altering the FEV1 value. In this model, the distal bronchioles also become infected due to the branching tree design of the airways (55-57). Formally, the equations are time delay differential equations (59) with switching at certain values of key variables, such as mucus volume and scarring.

To provide the best match between the model and clinical observations, values are determined for several parameters: the probability of onset of infection at any site, the rate of mucus buildup, the scarring rate, and the scarring threshold for onset of irreversible restructuring. In the simplest version of this model, treatments reset the mucus volume of the infected regions to zero unless scarring had exceeded the scarring threshold at the time of the treatment. Simulations can then be run and their predicted outcomes compared to the data for patient FEV1 responses available in
lung function databases. The model is then adjusted and the cycle repeated for several iterations (59).

The Community Dynamics Model: This approach has the challenging task of integrating several factors in order to predict clinically-significant changes in the microbial populations. These interacting factors include: 1) host immune responses and clinical therapies; 2) spatial heterogeneity of the CF airways; 3) the transitioning of aerobic microenvironments that support the rapidly-growing planktonic Attack community to anaerobic niches populated by slowly growing Climax populations within biofilms; and 4) phage predation. Accurate modeling of advanced, chronically-infected airways of CF patients will require validated submodels that represent each of these four factors. Currently, we lack sufficient empirical data to develop and test a comprehensive model. However, we can approach the modeling of phage predation using models already developed for predator-prey interactions, and this submodel can serve as an example. The populations of predators (e.g., phage) and prey (e.g., bacteria) are often observed to vary cyclically. An increased prey population supports more predators; more predators reduce the number of prey; fewer prey cause a crash in the predator population that allows the prey population to rebound. These dynamic fluctuations have been simulated for phage and bacteria using generalized Lotka-Volterra modeling based on a pair of linear differential equations. Applying this to the CF lung is complicated by local variation in the concentrations of phage or bacteria due to the microbial-scale spatial heterogeneity, as well as variation over time due to the effects of the immune system and clinical therapies.

Implications for Clinical Treatment: Each of the three modeling approaches described above quantitatively describes important aspects of the disease progression in CF patients. The models are inter-related in that one predicts values for parameters used by another. Together they provide a valuable adjunct for clinical treatment. It is neither possible nor desirable to perform all available treatments, in various combinations and each with varied timing of administration, on all patients. When a CF patient presents, a physician must recommend a treatment protocol, perhaps choosing
one antibiotic from among five possibilities, each with different treatment schedules. This is where the models become particularly valuable, because essentially all possibilities can be explored by computer simulations to predict which regimen would be most likely to increase patient FEV1 most quickly.

However, the ability of these models to describe the CF lung ecosystem and predict its behavior depends on the assignment of verified values to the parameters involved. To that end, we suggest that acquisition of the following data for airways and tissues of the CF lung are essential: 1) population dynamics for the microbial communities; 2) the distribution of microbial taxonomic and metabolic diversity across space and time; 3) nutrient sources for the microbial communities; 4) growth rates of specific microbes; 5) the types and numbers of viruses; and 6) the rates of phage-mediated mortality of microbes. With values for these variables, it will be possible to model the CF lung microbial-viral community using a variety of standard and proposed ecological models (Zarei, submitted).

Microbial ecologists have already developed the methods needed to assign values for these parameters. The next step is to apply them to the CF lung. Initial studies using metagenomic analysis of lung microbial communities have been reported (1, 3, 5, 15, 17, 18). Continuing investigations can provide critical information on the population dynamics and community structure of the microbial airway communities in health and disease. There is a reasonable expectation that such studies may identify an as-yet-unrecognized Achilles' heel within the pathogenic microbial populations in CF lungs. Metagenomic studies also can provide information on the natural history of the viral communities in the CF lung, an essentially unexplored topic. Of particular relevance are the complex relationships between the bacteria and their phage predators, as well as the possibility that viruses pathogenic to humans may be associated with the progression of lung disease.

**Summary**
The recent methodological developments that spurred rapid advancements in microbial ecology also provide new opportunities for understanding and controlling disease states in the CF lung. We propose an ecological model of the diseased lung that identifies two interrelated microbial communities: the aggressive Attack community and the more stable Climax community. The ecological approach that we advocate considers the interplay of the CF airway microbial and viral communities. Modeling of these interactions has the potential to predict the effects of therapeutic interventions, thus dispelling much of the current therapeutic empiricism and leading to more effective use of existing CF therapies. Lastly, employing principles of restoration ecology may lead to new therapeutic approaches designed to shift the CF airway communities to a less pathogenic state.
Acknowledgements: The authors would like to thank Angela Wang and Mike Furlan for their critical reading and comments on the manuscript text and Sean Needham for the medical illustration.
References


18(2):135-49.

55. Kitaoka, H., and B. Suki. 1997. Branching design of the bronchial tree based on a diameter-

structure affects lung blood flow heterogeneity simulated in three dimensions. *J Appl Physiol*
83(4):1370-82.

57. Weibel, E. 1991. Design of airways and blood vessels considered as branching trees. The

bifurcations of vector fields. Springer-Verlag, New York.

Thomson Brooks/Cole.
**Definition Table** (In a box)

**Bottom-up population control:** The control of the abundance of organisms by limited resources (e.g., nutrients) or environmental factors (e.g., oxygen concentration).

**Landscape ecology:** The study of the spatial heterogeneity of biotic and abiotic factors in an ecosystem, focusing on the accompanying interactions and their consequences for ecological processes.

**Metagenomics:** The culture-independent genomic analysis of an assemblage of microorganisms collected from an environment (e.g., the human gut, the CF lung). Phylogenetic typing discloses "who is there," while functional metagenomics reveals the metabolic capabilities of the entire sampled community, i.e., "what are they doing."

**Microbes:** Bacteria and Archaea. Although Archaea play important roles in biogeochemical cycles, none are known to be pathogens.

**Microbiota:** The microscopic organisms inhabiting a particular environment, including Bacteria, Archaea, viruses, and fungi.

**Phage (Bacteriophage):** A virus that infects Bacteria.
**Top-down population control:** The control of the abundance of organisms by predation. For the microbial populations in CF airways, top-down control is effected by bacteriophage and the immune system.
Figure 1.

Proposed Climax-Attack Model (CAM) for Cystic Fibrosis. The Attack communities consist of pathogens such as *Streptococcus* spp., *Staphylococcus* spp., and eukaryotic viruses. These communities elicit strong immune responses and scarring. The scar tissue is then colonized by the Climax community, which consists of the facultative anaerobe *P. aeruginosa* at the periphery and strict anaerobes in the central region.
Climax-Attack Model

**Attack Community**
- Environmentally acquired Bacteria
  - S. aureus
  - S. pneumonia
  - non-mucoid P. aeruginosa
  - H. influenza
- Endogeneous Bacteria
  - P. aeruginosa scv revertants
- Eukaryotic viruses
  - Rhinovirus
  - Adenovirus
  - Influenza A/B

**Climax Community**
- Bacteria
  - Mucoid P. aeruginosa
  - P. aeruginosa scv
  - Chronic S. aureus
  - S. maltophilia
  - Achromobacter spp.
  - Strict/Facultative anerobes
- Fungi
  - Aspergillus spp.
  - Scedosporium spp.

**Bronchiectasis**

**Hi pO2**

**Low pO2**