# Airway Bacterial Concentrations and Exacerbations of Chronic Obstructive Pulmonary Disease

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***Rationale*: Increased bacterial concentration (load) in the lower airways and new bacterial strain acquisition have been posited as mechanisms for chronic obstructive pulmonary disease (COPD) exacerbations. Bacterial concentrations are higher during exacerbation than during stable disease; however, these studies are cross sectional and devoid of strain typing.**

***Objectives*: To determine if the increased bacterial concentrations function as a separate mechanism for exacerbation induction independent of new strain acquisition.**

***Methods*: In a prospective, longitudinal cohort of patients with COPD, the relationship between exacerbation occurrence, sputum bacterial concentrations, and new strain acquisition was examined.**

***Measurements and Main Results*: Clinical information, quantitative sputum cultures, and molecular typing of potential bacterial pathogen isolates. Over 81 months, 104 subjects completed 3,009 clinic visits, 560 (19.6%) during exacerbations and 2,449 (80.4%) during stable disease. Among preexisting strains, sputum concentrations of Nontypeable *Haemophilus influenzae* and *Haemophilus haemolyticus* were not different in exacerbation versus stable disease. *Moraxella catarrhalis* (stable, $10^{8.38 \pm 0.13}$ [mean ± SEM] vs. exacerbation, $10^{7.78 \pm 0.26}$; $p = 0.02$) and *Streptococcus pneumoniae* (stable, $10^{8.42 \pm 0.21}$ vs. exacerbation, $10^{7.76 \pm 0.52}$; $p = 0.07$) concentrations were lower during exacerbations compared with stable periods. Concentrations of new strains of *H. influenzae* (stable, $10^{7.28 \pm 0.15}$ vs. exacerbation, $10^{7.76 \pm 0.17}$; $p = 0.04$) and *M. catarrhalis* (stable, $10^{7.85 \pm 0.15}$ vs. exacerbation, $10^{8.37 \pm 0.14}$; $p = 0.02$), were increased during exacerbations; however, the differences were small.**

***Conclusions*: Change in bacterial load is unlikely to be an important mechanism for exacerbations. Better understanding of the host–pathogen interaction, rather than enumerating bacteria in respiratory samples, is required to provide new insights into bacterial infection in COPD.**

**Keywords:** bacteria; chronic obstructive pulmonary disease; exacerbation

### AT A GLANCE COMMENTARY

#### Scientific Knowledge on the Subject

Bacterial concentrations in the lower airways are higher during exacerbations of chronic obstructive pulmonary disease; however, it is not known if the increased bacterial concentrations are independent of acquisition of new strains and function as a separate mechanism for exacerbation induction.

#### What This Study Adds to the Field

Sputum concentrations of preexisting bacterial strains were not higher during exacerbations. Among new strains, small increases were seen. These results demonstrate that change in bacterial load is unlikely to be an important mechanism for exacerbations of COPD.

Exacerbations are a significant contributor to the morbidity, mortality, health care costs, and impaired health status associated with chronic obstructive pulmonary disease (COPD) (1, 2). Bacterial etiology in about 50% of exacerbations is substantiated by several new lines of evidence, including bronchoscopic isolation of bacteria in the distal airways, the relationship of new strain isolation and exacerbations, the development of specific immune response to the infecting pathogen, as well as association of neutrophilic airway inflammation with bacterial isolation during exacerbations (3, 4).

Recently, new strain acquisition has been demonstrated as a mechanism of exacerbations; however, exacerbations are observed in the presence of preexisting bacterial strains—those isolated from the sputum before the onset of exacerbations (5). Furthermore, before the discovery of the relationship between new strain acquisition and exacerbations, the prevailing hypothesis of exacerbation mechanism was increased bacterial concentration (load) in the lower airways, with consequent increase in airway inflammation resulting in increased respiratory symptoms (6, 7). In fact, in several recent investigations, bacterial concentrations have been shown to be higher during exacerbation than during stable disease (7–9). However, many of these studies are cross sectional in nature, in which patients with exacerbations are compared with those with stable disease (8). In these studies, as well as in the studies that have employed longitudinal sampling, strain typing has not been performed (7–9). Therefore, from these studies, it is not possible to determine the relationship between increased bacterial concentrations observed at exacerbation and acquisition of new strains, and elucidate whether these are separate mechanisms for exacerbation induction.

We hypothesized that increased bacterial concentrations in the lower airway of patients with COPD is associated with exacerbation, independent of the acquisition of new strains of bacteria. In a prospective, longitudinal study of the dynamics of bacterial infection in COPD, quantitative bacterial cultures of sputum samples obtained from a cohort of patients with COPD were performed on a monthly basis, as well as during exacerbations. The relationship between exacerbation occurrence, bacterial

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concentrations in sputum, and acquisition of new strains was examined. The following hypotheses were tested: (*1*) Are bacterial concentrations in sputum related to the presence of exacerbation? (*2*) Are bacterial concentrations higher in exacerbation after accounting for the characterization of the strain as new or preexisting? If an increase in bacterial concentration is an independent mechanism for exacerbations, one would anticipate that there would be a relationship between higher bacterial concentrations and exacerbations among both new and preexisting strains. (*3*) Is there a relationship between bacterial concentrations and the occurrence of exacerbation after accounting for the acquisition of new strains as a confounding factor? Some of the results of these studies have been previously reported in the form of an abstract (10).

## METHODS

### COPD Study Clinic

The study protocol was approved by the Veterans Affairs Western New York Healthcare System institutional review board. This prospective cohort study has been described earlier in several publications (5, 11, 12). A total of 104 patients with COPD were enrolled between March 1994 and December 2000. The patients were seen monthly and whenever they had symptoms suggestive of an exacerbation. At each visit, clinical information, sputum, and serum samples were collected. Details of the study clinic are described in the online supplement.

### Quantification of Bacterial Pathogens in Sputum

Sputum sample processing and potential bacterial pathogen quantification is described in the online supplement. Potentially pathogenic bacteria included: *Haemophilus* spp., including Nontypeable *Haemophilus influenzae, H. haemolyticus*, and *H. parainfluenzae*, *Moraxella catarrhalis*, *Streptococcus pneumoniae*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and gram-negative enteric rods. Nontypeable *H. influenzae* is hereafter referred to as simply *H. influenzae*.

### Estimated Counts

Accurate colony counts could be determined from concentrations from $10^4$ to $10^9$ cfu/ml of homogenized sputum. Exact colony counts and, therefore, exact sputum concentrations were not available for potentially pathogenic bacteria in all sputum samples. The most common reason was that the potentially pathogenic bacterial colonies were mixed with normal flora and were, therefore, not discrete enough for accurate counts. Another reason was the presence of too many colonies to count on the highest dilution. Only rarely was the colony count data not recorded. For each of these instances, estimated counts of 1 log range were obtained as follows: for instances in which too many colonies were present at the highest dilution, bacterial colony count was estimated to lie between $1 \times 10^9$ and $1 \times 10^{10}$ cfu/ml. For the instances in which potentially pathogenic bacterial colonies were not discrete enough for accurate counts, bacterial colony count was estimated to lie between the 1 log range corresponding to 1 and 10 colonies for the plate with the highest dilution of sputum, where at least one colony was seen. For missing data, the same estimates described *above* were used.

In certain circumstances, neither exact nor estimated counts were available. This included many instances of isolation of *S. pneumoniae*, when present at relatively low concentrations, because of the colony morphology resemblance between this pathogen and colonies of commensal α-streptococci. In addition, colony counts were not performed for gram-negative bacilli (including *Pseudomonas*) and *Staphylococcus* spp.

### Molecular Typing

Molecular typing of strains of *H. influenzae*, *S. pneumoniae*, and *M. catarrhalis* was performed as described previously (5). Each strain was categorized as old or new based on molecular typing. A new strain was a strain that had not been previously isolated from sputum samples obtained from previous visits since the patient's enrollment. An old strain was one that had been isolated from sputum obtained from previous visits since the patient's enrollment. On further characterization of *H. influenzae* strains, we found that a substantial proportion were actually *H. haemolyticus*, many of which were nonhemolytic (13). These strains were analyzed separately from the *H. influenzae* strains.

### Data Analysis

Two different methods of data analysis were used. In the first instance, all colony counts were pooled and concentrations compared for each species for new and preexisting strains, separately. Concentrations used in all analyses were logarithm transformed. For estimated counts, logarithm-transformed concentrations were calculated at a random point of the log interval estimate. Because of multiple isolations of individual strains of pathogens from individual patients, generalized estimating equations (GEEs) were used for this analysis (14). The outcome variable was the presence of an exacerbation. The input variables included bacterial concentrations and whether the strain was new or preexisting.

In a second analysis, instances in which the same strain was isolated repeatedly from the same patient were determined. Of these instances, isolation of the same strain from a patient with an exacerbation and during stable state was analyzed further with paired *t* tests. If more than one visit in the stable state or exacerbation state were described for an individual strain, average values for these visits were used.

## RESULTS

### Subjects and Clinic Visits

From 1994 to 2000, over 81 months, 104 subjects completed 3,009 clinic visits. The demographic and clinical characteristics of these subjects are described in Table 1. Of the 3,009 clinic visits, 560 (19.6%) were during exacerbations and 2,449 (80.4%) were during stable COPD. Table 2 shows the number of isolates for each of the potential pathogens isolated from March 1994 to December 2000, and the number of isolates for which titers (exact and estimated) were available and were included in this study. Isolates for which titers were excluded or not available were as follows: (*1*) isolates obtained during repeated visits for the same exacerbation: *H. influenzae*, 8; *H. haemolyticus*, 1; *S. pneumoniae*, 1; *M. catarrhalis*, 1; and (*2*) isolates for which adequate information was not available to obtain either an exact or estimated titer: *S. pneumoniae*, 33; *M. catarrhalis*, 6; *H. parainfluenzae*, 5.

**TABLE 1. DEMOGRAPHIC AND CLINICAL CHARACTERISTICS OF STUDY CLINIC SUBJECTS**

| Characteristics | Value |
|---|---|
| Mean age (range), yr | 67.1 (45–85) |
| Sex, n | |
| Male | 102 |
| Female | 2 |
| Race, n | |
| White | 87 |
| African American | 17 |
| Mean time since diagnosis (range), yr | 11.2 (0–54) |
| Smoking status on enrollment, n | |
| Ex-smokers | 70 |
| Current smokers | 34 |
| Mean smoking (range), pack-years | 82.6 (10–185) |
| Mean $FEV_1$, L (range) | 1.56 (0.47–4.07) |
| Mean $FEV_1$% predicted (range) | 46.0 (15–99) |
| GOLD severity, no. of subjects | |
| Chronic bronchitis only | 5 |
| 1 | 1 |
| 2 | 35 |
| 3 | 41 |
| 4 | 22 |

*Definition of abbreviation*: GOLD = Global Initiative for Chronic Obstructive Lung Disease.

GOLD severity is based on global initiative for chronic obstructive lung disease guidelines (26).

**TABLE 2. DESCRIPTION OF ISOLATES OF EACH POTENTIAL RESPIRATORY PATHOGEN INCLUDED IN THIS STUDY**

| Pathogen | No. of Isolates | No. of Isolates Included* | No. with Exact Concentrations | No. with Estimated Concentrations |
|---|---|---|---|---|
| HI | 375 | 367 | 262 | 105 |
| HH | 190 | 189 | 150 | 39 |
| SP | 90 | 56 | 38 | 18 |
| MC | 194 | 187 | 140 | 47 |
| HP | 1,551 | 1,546 | 1,330 | 216 |

*Definition of abbreviations*: HH = *Haemophilus haemolyticus*; HI = nontypeable *Haemophilus influenzae*; HP = *Haemophilus parainfluenzae*; MC = *Moraxella catarrhalis*; SP = *Streptococcus pneumoniae*.

* Reasons for exclusion are provided in the text.

### Species Differences in Bacterial Titers

Different bacterial species may be present at different concentrations in the airway in COPD (Figure 1). This was determined by pair-wise comparisons of the mean concentrations among species. *S. pneumoniae* and *M. catarrhalis* exhibited the highest concentrations, significantly greater than all the other bacterial species, but equivalent to each other. Sputum concentrations of *H. haemolyticus* and *H. parainfluenzae* were significantly lower than all the other bacterial species, but equivalent to each other. Concentrations of *H. influenzae* were significantly higher than those of the other two *Haemophilus* species.

### Bacterial Titers, New Strains, and Clinical Status

Generalized estimating equations were constructed for each of the following bacterial pathogens: *H. influenzae*, *H. haemolyticus*, *H. parainfluenzae, S. pneumoniae*, and *M. catarrhalis*. Strain typing information was available for all these pathogens, with the exception of *H. parainfluenzae*.

### Bacterial Titers and Clinical Status

We first determined whether bacterial concentrations in sputum are related to the presence of exacerbation. If all strains, new and preexisting, were examined together, we found no significant difference between concentrations in sputum during stable disease and exacerbation for *H. influenzae*, *H. haemolyticus*, and *M. catarrhalis* (Figure 2). An inverse relationship between exacerbation occurrence and bacterial concentrations in sputum was seen for *S. pneumoniae* and *H. parainfluenzae*. Concentrations of these pathogens were actually lower during exacerbations than during stable disease (*S. pneumoniae*: stable, $10^{8.42 \pm 0.17}$ [mean ± SEM] vs. exacerbation, $10^{7.62 \pm 0.36}$, p = 0.048; *H .parainfluenzae*: stable, $10^{6.43 \pm 0.02}$ vs. exacerbation, $10^{6.29 \pm 0.05}$; p = 0.02), although the differences between exacerbation and stable visits were less than a log (10-fold).

### Bacterial Titers, Strain Acquisition, and Exacerbations

To test the major hypothesis of this study, we then determined if bacterial concentrations were associated with the occurrence of exacerbation after controlling for new strain acquisition using GEE models. In one approach, concentrations in sputum during stable disease and exacerbation were compared separately for new and preexisting isolates. As shown in Table 3, among new strains of *H. influenzae* and *M. catarrhalis*, but not for *H. haemolyticus* or *S. pneumoniae*, increased concentrations were indeed seen during exacerbation compared with during stable visits (*H. influenzae*: stable, $10^{7.28 \pm 0.15}$ vs. exacerbation, $10^{7.76 \pm 0.17}$; p = 0.04; *M. catarrhalis*: stable, $10^{7.85 \pm 0.15}$ vs. exacerbation, $10^{8.37 \pm 0.14}$; p = 0.02). However, the difference in concentrations between stable and exacerbation visits in each instance was about 0.5 log.

Among preexisting strains, no differences in sputum concentrations of *H. influenzae* and *H. haemolyticus* were found during exacerbation compared with stable disease (Table 3). *M. catarrhalis* was actually present at significantly lower concentrations during exacerbations as compared with stable periods (stable, $10^{8.38 \pm 0.13}$ vs. exacerbation, $10^{7.78 \pm 0.26}$; p = 0.02), and a similar trend was seen for *S. pneumoniae* (stable, $10^{8.42 \pm 0.21}$ vs. exacerbation, $10^{7.76 \pm 0.52}$; p = 0.07) (Table 3). As observed earlier, all differences were again less than a log.

In a second analytic approach, a GEE model considering the occurrence of exacerbation as the outcome with concentration as independent variable and new strain acquisition as a confounder was determined for each pathogen. In these models, bacterial titers of *H. haemolyticus* (p = 0.80) and *M. catarrhalis* (p = 0.20) had no relationship to the occurrence of exacerbation. Higher concentrations of *H. influenzae* were associated with exacerbation after accounting for new strain acquisition (p = 0.03; odds ratio, 1.27; 95% confidence interval, 1.03–1.56), but,

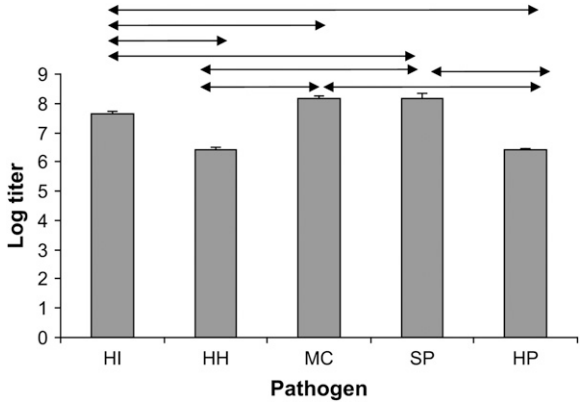


*Figure 1.* Sputum concentrations for each pathogen (mean ± SEM) in all sputum samples. *Double-headed arrows* denote significant differences (p < 0.05; generalized estimating equation with Tukey's adjustment for multiple comparisons). Pathogen: HH = *Haemophilus haemolyticus* (n = 189); HI = nontypeable *Haemophilus influenzae* (n = 367); HP = *Haemophilus parainfluenzae* (n = 1546); MC = *Moraxella catarrhalis* (n = 187); SP = *Streptococcus pneumoniae* (n = 56).

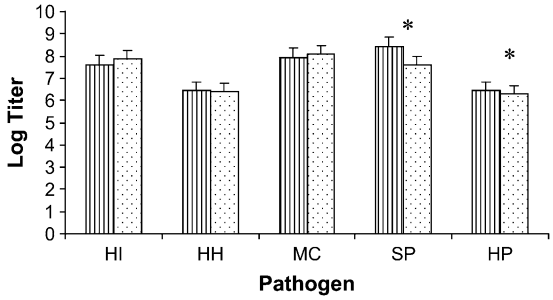


*Figure 2.* Bacterial concentration (mean ± SEM) in sputum for each pathogen in stable (*striped bars*) and exacerbation (*dotted bars*) visits. *Significant differences (p < 0.05). Pathogen: HH = *H. hemolyticus* (n = 158 stable, n = 31 exacerbation); HI = nontypeable *H. influenzae* (n = 288 stable, n = 79 exacerbation); HP = *H. parainfluenzae* (n = 1282 stable, n = 264 exacerbation); MC = *M. catarrhalis* (n = 120 stable, n = 67 exacerbation); SP = *S. pneumoniae* (n = 40 stable, n = 16 exacerbation).

**TABLE 3. SPUTUM BACTERIAL CONCENTRATIONS DURING EXACERBATION AND STABLE DISEASE FOR NEW AND PREEXISTING STRAINS**

| Pathogen/Clinical Status | No. of Strains | Mean Log Concentration (*SEM*) | p Value |
|---|---|---|---|
| New strains | | | |
| HI | | | |
| Stable | 78 | 7.28 (0.15) | 0.04 |
| Exacerbation | 44 | 7.76 (0.17) | |
| MC | | | |
| Stable | 63 | 7.85 (0.15) | 0.02 |
| Exacerbation | 51 | 8.37 (0.14) | |
| SP | | | |
| Stable | 16 | 8.32 (0.30) | 0.26 |
| Exacerbation | 7 | 7.53 (0.53) | |
| HH | | | |
| Stable | 82 | 6.42 (0.10) | 0.79 |
| Exacerbation | 21 | 6.38 (0.112) | |
| Preexisting strains | | | |
| HI | | | |
| Stable | 210 | 7.71 (0.09) | 0.31 |
| Exacerbation | 35 | 7.99 (0.20) | |
| MC | | | |
| Stable | 57 | 8.38 (0.13) | 0.02 |
| Exacerbation | 16 | 7.76 (0.26) | |
| SP | | | |
| Stable | 24 | 8.42 (0.21) | 0.07 |
| Exacerbation | 9 | 7.76 (0.52) | |
| HH | | | |
| Stable | 76 | 6.45 (0.10) | 0.98 |
| Exacerbation | 10 | 6.38 (0.29) | |

*Definition of abbreviations*: HH = *Haemophilus haemolyticus*; HI = nontypeable *Haemophilus influenzae*; MC = *Moraxella catarrhalis*; SP = *Streptococcus pneumoniae*.

**TABLE 4. PAIRED COMPARISON OF CONCENTRATIONS OF THE SAME STRAIN OF PATHOGENS ISOLATED FROM THE SAME PATIENT DURING EXACERBATION AND STABLE VISITS**

| Pathogen/Clinical Status | No. of Strains | Mean Concentration (*SEM*) | p Value |
|---|---|---|---|
| HI | | | |
| Stable | 34 | 7.51 (0.19) | 0.02 |
| Exacerbation | 34 | 8.08 (0.19) | |
| MC | | | |
| Stable | 15 | 7.79 (0.28) | 0.67 |
| Exacerbation | 15 | 7.89 (0.29) | |
| SP | | | |
| Stable | 7 | 8.81 (0.34) | 0.14 |
| Exacerbation | 7 | 8.50 (0.33) | |
| HH | | | |
| Stable | 13 | 6.09 (0.15) | 0.16 |
| Exacerbation | 13 | 6.38 (0.16) | |

For definition of abbreviations, *see* Table 3.

for *S. pneumoniae*, lower concentrations were found at exacerbation (p = 0.02; odds ratio, 0.88; 95% confidence interval, 0.80–0.98). This analysis confirmed some of the findings of the GEE models described previously here.

These analyses could not be conducted for *H. parainfluenzae*, as strain typing has not been performed for this potential pathogen.

### Paired Analysis of Strains

In our longitudinal collection of samples, we often encountered a strain that would be isolated during exacerbations and during stable visits from the same patient. These instances were an even more rigorous test of the hypothesis that bacterial concentrations are associated with exacerbations, as they represent a change in clinical status while the host and pathogen apparently remain the same. Results shown in Table 4 demonstrate that only *H. influenzae* (stable, $10^{7.51 \pm 0.19}$ vs. exacerbation, $10^{8.08 \pm 0.19}$; p = 0.02) was isolated at about 0.5 log higher concentration during exacerbation than during stable disease, with a trend toward lower concentrations during exacerbation for *S. pneumoniae*, and no differences observed for *M. catarrhalis* and *H. haemolyticus*. This analysis confirmed some of the findings of the GEE models described previously here.

## DISCUSSION

If increases in bacterial concentration (or load) in the lower airway was an independent mechanism of inducing exacerbations, we should have found increased bacterial concentrations during exacerbation when compared with stable disease among the preexisting strains (5). On the contrary, we found either no difference or, in some instances, even lower concentrations of these preexisting strains during exacerbation, demonstrating that change in bacterial load is unlikely to be an important mechanism for exacerbations. One can speculate that these exacerbations are likely caused by other pathogens (viruses or atypical bacteria) or environmental stimuli, and the resultant inflammatory response nonspecifically inhibits the preexisting strains and, therefore, is responsible for lowering their concentration (15).

Increased concentrations of *H. influenzae* and *M. catarrhalis* were seen during exacerbations associated with new strains in comparison to when these new strains were causing colonization. However, this difference in bacterial load, and similar differences observed in other studies, although statistically significant, is unlikely to be of enough magnitude to be biologically significant (9). These differences are within 1 log (10-fold), whereas the total bacterial load in the airways is in the order of 7–8 logs. Therefore, these differences are actually quite small; for example, a 0.5 log difference is 7% of the total bacterial load (16). It is likely that these differences in bacterial load reflect the outcome of the host–pathogen interaction rather than an independent mechanism of exacerbation.

The strengths of this study are inclusion of molecular tying and longitudinal sampling. Molecular typing allowed discrimination of new strains from preexisting strains, as well as providing us with the ability to account for new strain acquisition when examining the relationship of bacterial concentrations to exacerbation. Our results were different from those of previous studies, which have found increased bacterial concentrations during exacerbations of COPD as compared with stable disease, but did not include molecular typing of bacterial strains, demonstrating the importance of accounting for strain acquisition (7–9). Longitudinal sampling in this study meant that the populations of patients contributing exacerbation and stable samples were the same. Therefore, potential confounding by different baseline characteristics of the populations being compared in previous studies with cross-sectional design was avoided (8). The paired analysis of patients when they have had the same strain isolated during exacerbation and stable disease is especially powerful in controlling for confounding variables, and did not demonstrate an effect of bacterial load, with the exception of a small difference in the case of *H. influenzae*.

We did find differences in concentrations of pathogens among species. Pathogens with the most clearly established role in exacerbations of COPD, *H. influenzae*, *S. pneumoniae*, and *M. catarrhalis*, were present in higher concentrations compared with

pathogens that are more likely to be colonizers (*H. parainfluenzae* and *H. haemolyticus*) (11–13, 17, 18). This may reflect differences in virulence and adaptation to the human respiratory tract among the pathogenic species. These differences among potential pathogens should be taken in to account when interpreting studies examining bacterial concentrations and airway inflammation in COPD (7, 19).

Limitations of our study include the use of estimated concentrations in several instances instead of exact bacterial concentrations. However, our estimate is within a log of the actual concentration, and the statistical methods employed take in to account that these are estimates. Another limitation is that the bacterial concentrations were measured in sputum samples rather than bronchoscopically obtained samples, such as protected specimen brushings or bronchoalveolar lavage (8, 20, 21). The necessity for repeated sampling in our study design made sputum samples the only practical way to obtain these measurements. Our data are robust for *Haemophilus* spp. and *M. catarrhalis*; however, because of the limited number of observations with the pneumococcus, more observations are needed with this pathogen. In addition, other potential pathogens in exacerbations of COPD, such as *Pseudomonas aeruginosa* and *Staphylococcus aureus*, are not addressed in this study. The presence of viruses and atypical bacteria was not determined in this study. Such information could have added to the understanding of the mechanism of exacerbations, especially those with preexisting bacterial strains.

Studies of airway inflammation (unpublished observations) and immune response seem to support the concept that preexisting strains of the bacteria studied in this work are an infrequent cause of exacerbations (17). This study supports this concept, and, even if preexisting strains were to cause exacerbations, the mechanism does not appear to be an increase in bacterial load. Alternative mechanisms by which preexisting strains could cause exacerbations include alteration of their antigenic structure to evade the immune response or alteration of the airway milieu by another infection (e.g., a virus that alters the interaction between the host and the colonizing pathogen) (9, 16, 22). Such a process would be consistent with the multiple hit hypothesis recently proposed to explain variations in inflammation in airways diseases such as COPD (23). These mechanisms need investigation to further our understanding of host–pathogen interaction between colonizing bacterial strains and the host in COPD. However, this study does not preclude a pathogenic role for bacterial colonization in causing airway inflammation and inducing ongoing lung damage (19, 24, 25).

Are there clinical implications of the findings of this study? Quantitative cultures of sputum are rarely done in the clinical management of COPD. Semiquantitative sputum cultures are often performed and reported. Results of this study imply that these semiquantitative results are not useful in determining the etiologic role of the isolated pathogen. This study has obvious connotations for future research in this field. The host–pathogen interaction that underlies exacerbations of COPD is more complicated than simple changes in concentrations of bacteria. Studies limited to isolating and enumerating bacteria from respiratory samples are unlikely to provide us with new insights into bacterial infection in COPD. Better understanding of the host–pathogen interaction in COPD, including virulence factors of the bacterial pathogen and the alterations in the innate and adaptive lung defenses that allow bacteria to persist in the lower airway, are more likely to be fruitful.

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