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Age of *Pseudomonas aeruginosa* Acquisition and Subsequent Severity of Cystic Fibrosis Lung Disease

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Summary

Rationale: *Pseudomonas aeruginosa* (*Pa*) is associated with poor pulmonary outcomes in cystic fibrosis (CF), but the association between age of *Pa* infection and severity of subsequent lung disease has not been thoroughly investigated. **Objective:** Our goal was to determine the association between age of *Pa* acquisition and subsequent severity of CF lung disease. **Methods:** Case-control study using CF Foundation Registry data of 629 F508 homozygotes with severe and mild lung disease (FEV₁ in the lowest and highest quartile of birth cohort, respectively). Multivariate logistic regression was performed to determine the association between age of *Pa* acquisition and lung disease severity. **Results:** Earlier age of *Pa* infection was strongly associated with increased odds of severe lung disease. For first and persistent *Pa*, adjusted odds ratios for severe lung disease were 6.5 (95% CI 3.1, 13.7; *P* < 0.0001) and 11.2 (5.4, 23.1; *P* < 0.0001), respectively, for subjects with infection before age 5 versus at 10 years; the association was stronger in females than males. **Conclusions:** Earlier *Pa* infection, particularly before 5 years of age, is strongly associated with severe CF lung disease later in life. This study is not designed to determine causality; *Pa* infection may be causing lung injury, or may be a marker of ongoing inflammation and lung damage in young children with CF.

Keywords

pediatrics; infection; epidemiology; lung diseases; obstructive

INTRODUCTION

Pseudomonas aeruginosa (*Pa*) is a significant pathogen in cystic fibrosis (CF) lung disease^{1–3}; its prevalence in respiratory cultures increases with age, reaching approximately 80% by adulthood.⁴ Factors associated with acquisition of *Pa* in patients with CF include female gender, pancreatic exocrine insufficiency, viral infections (particularly respiratory syncytial virus), socioeconomic status, and previous infection with *Staphylococcus aureus* (*S. aureus*).^{5–9} Infection with *Pa*, particularly the mucoid strain,¹⁰ has been associated with decline in lung function and/or more severe lung disease, and earlier infection has been associated with lower forced expiratory volume in 1 sec (FEV₁) and higher risk of death in childhood.^{4,10–18} While there is considerable evidence supporting an association between *Pa* infection and more severe lung disease, previous studies have had several limitations, including small sample size, limited follow-up time, inclusion of subjects with varied CFTR genotype, and a multivariable analysis not designed to specifically assess the relationship between *Pa* infection and lung disease.

Our objective was to determine the association between age of *Pa* infection in childhood and severity of CF lung disease later in life in a large population of F508 homozygotes with an extended duration of respiratory culture data. Some of the results of this study have been previously reported in the form of an abstract.¹⁹

METHODS

Objective and Design

We performed a case–control study of subjects with CF enrolled in the University of North Carolina and Case Western Reserve University Gene Modifier Study (GMS),²⁰ which was designed to study CF modifier genes employing an extremes-of-phenotype design, with subjects with *severe* and *mild* lung disease serving as cases and controls, respectively. The University of North Carolina at Chapel Hill Institutional Review Board approved the current study; the GMS study was approved by the human studies or ethical review board at each participating institution, and informed consent was obtained from all subjects prior to enrollment.

Population

Patients enrolled in the GMS were F508 homozygotes and had either (a) lung function (FEV₁) in the lowest quartile for birth cohort and were 8–25 years old or (b) lung function in the highest quartile and were age 15. Siblings and subjects post lung transplantation or lobectomy were excluded from the GMS.²⁰ To avoid confounding by changes in treatment practices over time, we included only “young mild” (enrolled at age 15–28, n = 403) and severe subjects (enrolled at age 8–25, n = 406) who were enrolled in the GMS between January, 2000 and March, 2007.²⁰ Our analysis was restricted to the 629 subjects for whom

CF Foundation Patient Registry (CFFPR) data was available and health insurance status was known (Fig. 1).

Data

Data was obtained from the GMS [from which disease severity, age of GMS enrollment, and transforming growth factor- β (TGF- β) codon 10 genotype were assigned] and the CFFPR (all other data, including standardized clinical, demographic, and respiratory culture data).^{20,21}

Variable Definitions

Age of *Pa* infection (primary independent variable) was defined in two ways: (1) age of first *Pa*-positive culture, and (2) age of persistent *Pa*, defined as the age of the first of three consecutive years with *Pa*-positive cultures in at least 2 of the 3 years. Both variables were coded in 5 year blocks: *Pa* infection at age 0–4 years, 5–9 years, and the referent group consisting of those with infection at 10 years, or who never met *Pa*-infection criteria. Our outcome of interest was severity of CF lung disease in childhood and young adulthood (age 8–28) coded as severe or mild (Fig. 1).²⁰

Potential confounders included age of first respiratory culture data, years of culture data, birth year, age of CF diagnosis, TGF- β codon 10 genotype, gender, symptoms at diagnosis (respiratory and gastrointestinal vs. other), *S. aureus* infection (coded 3 ways: age of first *S. aureus* infection, age of persistent *S. aureus* infection, and *S. aureus* before or after first *Pa*), health insurance status (public vs. private), and CF center at time of CFFPR enrollment. Because of the potential for confounding by differences in management practices, CF centers (46 in total) were classified based on the centers' proportion of severe and mild subjects enrolled in the GMS [$>60\%$ severe; $>60\%$ mild; or even (referent)]. All time and age variables were coded in years.

Analysis

Bivariate analyses were performed using Student's *t*-tests and χ^2 -tests, as appropriate, with a *P*-value < 0.05 considered significant.²² Formal analysis was performed using multivariate logistic regression including potential confounders (listed above), identified *a priori* based on literature review, in the initial model.²² Analysis of the association of age of *Pa* infection with severity of disease was performed using age of *Pa* infection as both a continuous and categorical variable. For categorical coding, both 3 and 5 year blocks were tested, with cutoff for the referent group ranging from 10 to 15 years for age of *Pa* infection. The most precise classification (based on confidence limit ratio) was chosen for use in the final analysis (data not shown).

When testing for effect measure modification, a change in estimate (odds ratio; OR) of $>10\%$ was considered significant. Backwards elimination was employed to determine variables (potential confounders) included in the final model, with a change in estimate of $>10\%$ considered significant. The same rule was applied to sensitivity analyses to determine if changes in variable definition for CF center and symptoms at presentation affected

strength of the observed association between age of *Pa* and lung disease severity. All analyses were performed using Stata[®] version 10 (www.stata.com).

RESULTS

Study Population and Demographics

Final analysis included 310 subjects with severe disease and 319 with mild disease (Fig. 1). Summary data for severe and mild subject groups are shown in Table 1. There was no significant difference in gender distribution between the two groups. Subjects with severe lung disease were born later, diagnosed at a younger age, had fewer years of culture data, and enrolled in the GMS at a younger age than those with mild disease, likely all secondary to study design (enrollment ages).

Age of *Pseudomonas aeruginosa* and Severity of Lung Disease

Mean Age of *Pa* Infection (Bivariate Analysis)—Infection with *Pa* occurred approximately 5 years earlier in severe subjects than mild subjects, whether comparing age of initial or persistent *Pa* infection ($P < 0.0001$ for both; Table 1). Mild subjects were also more likely than severe subjects to never have persistent *Pa* infection (15% and 8% of subjects, respectively; $P < 0.0001$). The average time from first culture data to first *Pa*-positive culture was 2.8 versus 5.6 years in the severe versus mild group ($P < 0.0001$; Table 1, Fig. 2). Figure 2 plots the cumulative percentage of subjects with acquisition *Pa* by age, highlighting the pattern of earlier *Pa* infection in subjects with severe lung disease. Mean age of transition from oropharyngeal (OP) to sputum cultures—defined as the age at the time of the first of three consecutive years with 1 sputum culture reported in at least 2 of the 3 years—was significantly younger in severe than in mild subjects (7.6 vs. 11.7 years of age, respectively; $P < 0.0001$).

Age of *Pa* Infection as a Categorical Variable (Multivariate Analysis)—In developing a multivariate model of the effect of age of *Pa* acquisition on lung disease severity, we evaluated different classifications of the primary independent variable (age of *Pa* infection). We compared age of *Pa* infection as a continuous variable and as a categorical variable in 3 or 5 year blocks with the referent group composed of subjects with age of *Pa* infection at 10, 12, or 15 years. Similar patterns of association between age of *Pa* infection and severity of lung disease were seen regardless of the classification scheme employed (data not shown). Our final classification of *Pa* infection in 5 year blocks, with subjects with *Pa* infection at 10 years of age (or who were never *Pa* positive) as the referent group was chosen to obtain the most precise (smallest confidence limit ratio) estimate of association, and to allow for nonlinearity (compared to classification of age of *Pa* infection as a continuous variable) of the relationship between age of *Pa* infection and disease severity. Because a small percentage of subjects never met criteria for first or persistent *Pa* infection, these subjects were included in the referent group (see Table 1).

When age of *Pa* infection was treated as a categorical variable in 5 year blocks, there was a strong association between age of first *Pa* infection and severity of lung disease later in life both in crude analysis and when adjusted for years of culture data, age of first culture, and

health insurance status, which served as our final model (Table 2). Subjects with a first *Pa*-positive culture at 0–4 years of age had 5.9 (95% CI 3.2, 10.9; $P < 0.0001$) times the odds of severe disease compared to the referent group (those with first *Pa* at 10 years, or never *Pa* positive); subjects with their first *Pa*-positive culture at 5–9 years had 3.5 (95% CI 2.1, 5.8; $P < 0.0001$) times the odds of severe disease. An even stronger association was seen when comparing age of persistent *Pa* infection. Subjects with persistent *Pa* at 0–4 years of age had 11 (95% CI 5.5, 22.1; $P < 0.0001$) times the odds of severe disease as the referent group; those with persistent *Pa* at 5–9 years had 4.3 (95% CI 2.7, 6.8; $P < 0.0001$) times the odds of severe disease. *S. aureus* infection did not have a significant impact on the observed association between age of *Pa* and severity of CF lung disease (data not shown). Alternate definitions (sensitivity analyses) for CF center and symptoms at diagnosis did not have a significant impact on the association of interest.

Age of *Pa* Infection by Culture Type—A similar pattern of association between age of *Pa* infection and severity of CF lung disease was seen when analysis was restricted by culture type (years with only OP culture data and years with at least one sputum culture reported; data not shown). Specifically, subjects with severe disease had earlier onset (approximately 5–6 years) of *Pa* infection when analysis was restricted to either years with only OP culture data or years with at least one sputum culture reported (sputum culture data only or OP and sputum culture data available).

Impact of Gender on Association (Effect Measure Modification)

Because female gender has been associated with poorer pulmonary outcomes in CF,^{5–7,12,14,21,23} we assessed for a possible interaction effect (effect measure modification) of gender on the association between age of *Pa* infection and severity of lung disease (Table 3). The association between age of persistent *Pa* infection and severity of lung disease was significantly stronger in females than in males, despite the fact that there was no significant difference in mean age of diagnosis between females and males [1.75 (± 3.4) and 1.73 (± 3.5) years of age, respectively; $P = 0.9$]. The adjusted ORs for severe lung disease for subjects with persistent *Pa* between 0 and 4 years of age and between 5 and 9 years of age were approximately 9 times higher in females than in males, both compared to the same-sex referent group ($P = 0.003$). A similar pattern was seen when comparing gender differences in the association between age at first *Pa* infection and lung disease severity (Table 3).

DISCUSSION

This case-control study of 629 F508 homozygotes collected for the GMS shows a strong association between earlier age of acquisition of *Pa* and severity of CF lung disease later in life, even when adjusting for potential confounders. Subjects first culture-positive for *Pa* before 5 years of age had 6.5 times the odds of severe lung disease later in life compared to those who were first positive at 10 years or older (or who were never *Pa* positive); this association was even more pronounced for persistent *Pa*, where the odds of severe disease were 11.2 times higher in subjects with persistent *Pa* at 0–4 years of age compared to the referent group. There appeared to be a stronger association between age of *Pa* infection and likelihood of severe lung disease in females compared to males.

Though several previous studies have linked respiratory infection with *Pa* to poor pulmonary outcomes in CF, this study is one of the first to focus on specific age of infection, particularly in early childhood, in a large study population with an extended time span of respiratory culture data. We have minimized the impact of CFTR genetics by restricting subjects to F508 homozygotes. We also minimized potential confounding by birth cohort (from differences in treatment practices) by limiting our analysis to severe subjects and those “young milds” enrolled in the GMS between 15 and 28 years of age.²⁰ The majority of our subjects were first infected with *Pa* prior to the common use of inhaled tobramycin in pediatric CF clinics and in eradication protocols,²⁴ further lessening the impact of changes in treatment practices on our results.

In order to address the variability of respiratory culture data, we tested two definitions of *Pa* infection—first *Pa* and persistent *Pa*. Because existing classification systems for *Pa* infection rely on multiple points of culture data in a single year,^{25,26} we created a definition of persistent *Pa* infection to accommodate annualized data; specifically, age of persistent *Pa* was defined as the age of the first of three consecutive years with *Pa*-positive cultures in at least 2 of the 3 years. The observed association between age of *Pa* and severity of CF lung disease was present when either definition of *Pa* infection was used in the analysis, though a stronger association was seen when comparing age of persistent *Pa* and lung disease severity. Our multiple sensitivity analyses on characterization of age of *Pa* infection, including continuous coding, varying size and number of age blocks for categorical coding, and comparison of age of *Pa* infection by years with only OP culture data versus years with sputum culture data available, all yielded similar patterns of association, strengthening the likelihood that our results reveal a true association. Shorter time from first culture to first *Pa*-positive culture among subjects with severe versus mild lung disease (Table 1) indicates that the earlier age of *Pa* infection observed in the severe group is not simply due to earlier age of available culture data, but instead reflects a true predilection for earlier *Pa* acquisition. Sensitivity analyses of variable definitions for confounders did not have a significant impact on our results. The observed association implies that presence of *Pa* under age 10, and particularly before age 5, is strongly tied to poorer pulmonary outcomes in subjects with CF, but does not infer causality.

We saw an interaction between gender and age of *Pa* infection, with a stronger association between age of *Pa* infection and likelihood of severe lung disease in females compared to males (Table 3). This is consistent with previous reports of female gender as a risk factor for worse pulmonary outcomes in CF.^{5–7,12,14,23} These results suggest that females may deal more poorly with *Pa* infection, or that females with worse lung disease may be more prone to *Pa* infection than males with the same degree of disease.²⁷ The interaction between gender and age of *Pa* infection has yet to be fully determined, and bears further investigation in future studies.

Our findings are consistent with previous studies in subjects with CF which have shown *Pa* infection to be associated with increased endobronchial inflammation, poorer clinical status, more rapid progression of lung disease, and increased morbidity and mortality in young children with CF.^{2,3,11–13,18} Previous studies have also raised the question of whether *Pa* infection is a cause of decline in lung function or a marker of more severe disease.¹¹ Female

gender has also been shown in multiple studies to be an independent risk factor for more severe CF lung disease, similar to the findings in our study.^{5,14,23,28–30} Newer studies have suggested that *Pa* may be acquired in the preschool age range in the majority of patients with CF^{1,31}; this may be due to changes in culture practices as the CF community moved toward adopting eradication protocols.

We recognize our study has certain limitations, the most important of which is that we cannot determine causality based on our study design. Another limitation is the extremes of phenotype model, which limits our ability to generalize our results to the CF population as a whole. Our reliance on annualized data, made necessary by the later (1998) adoption of quarterly data into the CFFPR, means we cannot determine the density of *Pa*-positive cultures within a particular year, nor can we distinguish chronic infection from consecutive “new” episodes of *Pa* infection; however, we employed two distinct definitions of *Pa* infection with similar results. Staggered entry and exit times for CFFPR data have the potential to create bias (potentially leading to earlier age of *Pa* infection in those subjects with data at an earlier age), though the shorter time to *Pa* positivity seen in subjects with severe disease suggests that the observed earlier age of *Pa* infection in this group is not simply due to CFFPR data availability at a younger age. Staggered entry data in the CFFPR also limited our ability to determine and control for nutritional status early in life. Our database lacked strong indicators of socioeconomic status such as parental income, though we did employ health insurance status at the time of CFFPR enrollment as a proxy.⁹ We were unable to adjust for methicillin-resistant *S. aureus* or analyze the impact of mucoid versus non-mucoid *Pa* because these data were not collected until much later in our CFFPR dataset. We did not control for race/ethnicity, as less than 3% (n = 15) of our subjects were identified as non-Caucasian. False positives that are unrelated to the severity of disease, false negatives, and errors in reporting or recording of *Pa* status are a possibility in this study. However, such non-differential measurement errors would serve to attenuate the observed association between age of *Pa* infection and severity of CF lung disease in relation to the true population association, again suggesting that our reported association is a true one. Because the majority of our subjects acquired *Pa* prior to the routine use of inhaled tobramycin, our findings may not directly apply to current patients with CF. Despite these limitations, we believe that the robust association seen within our population between age of *Pa* infection and severity of later CF lung disease describes a true relationship, particularly since the association holds through multiple sensitivity analyses.

In summary, earlier age of infection with *Pa* in our population was strongly associated with greater likelihood of severe lung disease later in life, most particularly in those subjects who acquired *Pa* before age 5; the observed association was stronger in females than in males. *Pa* infection may be a cause of more severe CF lung disease, but may also be a marker of some other processes determining lung function in children with CF.

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ABBREVIATIONS

CF	Cystic fibrosis
CTFR	CF Transmembrane conductance regulator
<i>Pa</i>	<i>Pseudomonas aeruginosa</i>
<i>S. aureus</i>	<i>Staphylococcus aureus</i>
FEV₁	Forced expiratory volume in 1 sec
GMS	Gene Modifier Study
CFFPR	CF Foundation Patient Registry
TGF-β	Transforming growth factor- β
OR	Odds ratio
OP	Oropharyngeal

References

1. Burns JL, Gibson RL, McNamara S, Yim D, Emerson J, Rosenfeld M, Hiatt P, McCoy K, Castile R, Smith AL, Ramsey BW. Longitudinal assessment of *Pseudomonas aeruginosa* in young children with cystic fibrosis. *J Infect Dis*. 2001; 183:444–452. [PubMed: 11133376]
2. Nixon GM, Armstrong DS, Carzino R, Carlin JB, Olinsky A, Robertson CF, Grimwood K. Clinical outcome after early *Pseudomonas aeruginosa* infection in cystic fibrosis. *J Pediatr*. 2001; 138:699–704. [PubMed: 11343046]
3. Kosorok MR, Zeng L, West SE, Rock MJ, Splaingard ML, Laxova A, Green CG, Collins J, Farrell PM. Acceleration of lung disease in children with cystic fibrosis after *Pseudomonas aeruginosa* acquisition. *Pediatr Pulmonol*. 2001; 32:277–287. [PubMed: 11568988]
4. Rosenfeld M, Ramsey BW, Gibson RL. *Pseudomonas* acquisition in young patients with cystic fibrosis: Pathophysiology, diagnosis, and management. *Curr Opin Pulm Med*. 2003; 9:492–497. [PubMed: 14534401]
5. Demko CA, Byard PJ, Davis PB. Gender differences in cystic fibrosis: *Pseudomonas aeruginosa* infection. *J Clin Epidemiol*. 1995; 48:1041–1049. [PubMed: 7775991]
6. Levy H, Kalish LA, Cannon CL, Garcia KC, Gerard C, Goldmann D, Pier GB, Weiss ST, Colin AA. Predictors of mucoid *Pseudomonas* colonization in cystic fibrosis patients. *Pediatr Pulmonol*. 2008; 43:463–471. [PubMed: 18361452]
7. Maselli JH, Sontag MK, Norris JM, MacKenzie T, Wagener JS, Accurso FJ. Risk factors for initial acquisition of *Pseudomonas aeruginosa* in children with cystic fibrosis identified by newborn screening. *Pediatr Pulmonol*. 2003; 35:257–262. [PubMed: 12629621]
8. Petersen NT, Hoiby N, Mordhorst CH, Lind K, Flensburg EW, Bruun B. Respiratory infections in cystic fibrosis patients caused by virus, chlamydia and mycoplasma—Possible synergism with *Pseudomonas aeruginosa*. *Acta Paediatr Scand*. 1981; 70:623–628. [PubMed: 6798822]

9. Schechter MS, Shelton BJ, Margolis PA, Fitzsimmons SC. The association of socioeconomic status with outcomes in cystic fibrosis patients in the united states. *Am J Respir Crit Care Med*. 2001; 163:1331–1337. [PubMed: 11371397]
10. Li Z, Kosorok MR, Farrell PM, Laxova A, West SE, Green CG, Collins J, Rock MJ, Splaingard ML. Longitudinal development of mucoid *Pseudomonas aeruginosa* infection and lung disease progression in children with cystic fibrosis. *JAMA*. 2005; 293:581–588. [PubMed: 15687313]
11. Aebi C, Bracher R, Liechti-Gallati S, Tschappeler H, Rudeberg A, Kraemer R. The age at onset of chronic *Pseudomonas aeruginosa* colonization in cystic fibrosis—Prognostic significance. *Eur J Pediatr*. 1995; 154:S69–S73. [PubMed: 8529715]
12. Emerson J, Rosenfeld M, McNamara S, Ramsey B, Gibson RL. *Pseudomonas aeruginosa* and other predictors of mortality and morbidity in young children with cystic fibrosis. *Pediatr Pulmonol*. 2002; 34:91–100. [PubMed: 12112774]
13. Farrell PM, Shen G, Splaingard M, Colby CE, Laxova A, Kosorok MR, Rock MJ, Mischler EH. Acquisition of *Pseudomonas aeruginosa* in children with cystic fibrosis. *Pediatrics*. 1997; 100:E2. [PubMed: 9346996]
14. Konstan MW, Morgan WJ, Butler SM, Pasta DJ, Craib ML, Silva SJ, Stokes DC, Wohl ME, Wagener JS, Regelman WE, Johnson CA. Risk factors for rate of decline in forced expiratory volume in one second in children and adolescents with cystic fibrosis. *J Pediatr*. 2007; 151:134–139. [PubMed: 17643762]
15. Schaedel C, de Monestrol I, Hjelte L, Johannesson M, Kornfalt R, Lindblad A, Strandvik B, Wahlgren L, Holmberg L. Predictors of deterioration of lung function in cystic fibrosis. *Pediatr Pulmonol*. 2002; 33:483–491. [PubMed: 12001283]
16. Gomez MI, Prince A. Opportunistic infections in lung disease: *Pseudomonas* infections in cystic fibrosis. *Curr Opin Pharmacol*. 2007; 7:244–251. [PubMed: 17418640]
17. Kozłowska WJ, Bush A, Wade A, Aurora P, Carr SB, Castle RA, Hoo AF, Lum S, Price J, Ranganathan S, Saunders C, Stanojevic S, Stroobant J, Wallis C, Stocks J. Lung function from infancy to the preschool years following clinical diagnosis of cystic fibrosis. *Am J Respir Crit Care Med*. 2008; 178:42–49. [PubMed: 18403721]
18. Sagel SD, Gibson RL, Emerson J, McNamara S, Burns JL, Wagener JS, Ramsey BW. Impact of *Pseudomonas* and *Staphylococcus* infection on inflammation and clinical status in young children with cystic fibrosis. *J Pediatr*. 2009; 154:183–188. [PubMed: 18822427]
19. Pittman JCE, Yeatts J, Drumm M, Leigh MW, Davis SD, Van Rie A, Emond M, Knowles M. Age of *Pseudomonas aeruginosa* infection and severity of lung disease in cystic fibrosis adolescents and adults. *Pediatr Pulmonol*. 2008; A332:319. Abstracts edition.
20. Drumm ML, Konstan MW, Schluchter MD, Handler A, Pace R, Zou F, Zariwala M, Fargo D, Xu A, Dunn JM, Darrah RJ, Dorfman R, Sandford AJ, Corey M, Zielenski J, Durie P, Goddard K, Yankaskas JR, Wright FA, Knowles MR. Genetic modifiers of lung disease in cystic fibrosis. *N Engl J Med*. 2005; 353:1443–1453. [PubMed: 16207846]
21. FitzSimmons SC. The changing epidemiology of cystic fibrosis. *J Pediatr*. 1993; 122:1–9. [PubMed: 8419592]
22. Rothman, K.; Greenland, S., editors. *Modern epidemiology*. Philadelphia, PA: Lippincott Williams & Wilkins; 1998.
23. Davis PB. The gender gap in cystic fibrosis survival. *J Gend Specif Med*. 1999; 2:47–51. [PubMed: 11252861]
24. Ramsey BW, Pepe MS, Quan JM, Otto KL, Montgomery AB, Williams-Warren J, Vasiljev KM, Borowitz D, Bowman CM, Marshall BC, Marshall S, Smith AL. Intermittent administration of inhaled tobramycin in patients with cystic fibrosis. Cystic fibrosis inhaled tobramycin study group. *N Engl J Med*. 1999; 340:23–30. [PubMed: 9878641]
25. Proesmans M, Balinska-Miskiewicz W, Dupont L, Bossuyt X, Verhaegen J, Hoiby N, de Boeck K. Evaluating the “Leeds criteria” for *Pseudomonas aeruginosa* infection in a cystic fibrosis centre. *Eur Respir J*. 2006; 27:937–943. [PubMed: 16707392]
26. Lee TW, Brownlee KG, Conway SP, Denton M, Littlewood JM. Evaluation of a new definition for chronic *Pseudomonas aeruginosa* infection in cystic fibrosis patients. *J Cyst Fibros*. 2003; 2:29–34. [PubMed: 15463843]

27. Guilbault C, Stotland P, Lachance C, Tam M, Keller A, Thompson-Snipes L, Cowley E, Hamilton TA, Eidelman DH, Stevenson MM, Radzioch D. Influence of gender and interleukin-10 deficiency on the inflammatory response during lung infection with *Pseudomonas aeruginosa* in mice. *Immunology*. 2002; 107:297–305. [PubMed: 12423305]
28. Rosenfeld M, Davis R, FitzSimmons S, Pepe M, Ramsey B. Gender gap in cystic fibrosis mortality. *Am J Epidemiol*. 1997; 145:794–803. [PubMed: 9143209]
29. Verma N, Bush A, Buchdahl R. Is there still a gender gap in cystic fibrosis? *Chest*. 2005; 128:2824–2834. [PubMed: 16236961]
30. Corey M, Edwards L, Levison H, Knowles M. Longitudinal analysis of pulmonary function decline in patients with cystic fibrosis. *J Pediatr*. 1997; 131:809–814. [PubMed: 9427882]
31. West SE, Zeng L, Lee BL, Kosorok MR, Laxova A, Rock MJ, Splaingard MJ, Farrell PM. Respiratory infections with *Pseudomonas aeruginosa* in children with cystic fibrosis: Early detection by serology and assessment of risk factors. *JAMA*. 2002; 287:2958–2967. [PubMed: 12052125]

Fig. 1.

Subject enrollment criteria and diagram of study population. Subjects were included in our analysis if they (a) met Gene Modifier Study (GMS) enrollment criteria as either severe or young mild subjects, (b) had CF Foundation Patient Registry (CFFPR) data available, and (c*) health insurance status at the time of CFFPR enrollment was not missing or classified as “other,” “none,” or “unknown.” †One mild subject was excluded for lack of TGF- β genotype data.

Fig. 2.

Cumulative percentage of subjects ever reported positive for *Pseudomonas aeruginosa* (*Pa*) by age and lung disease severity. Subjects were considered *Pa* positive at a given age if they had a history of at least one culture positive for *Pa* by that age.

TABLE 1

Demographics and Culture Data

	Severe (n = 310) (SD)	Mild (n = 319) (SD)
Demographics		
Female	49%	46%
Year of birth	1987 (5.1) ¹	1982 (4.3)
Age (years) at CF diagnosis	1.3 (2.6) ²	2.1 (4.1)
Age (years) at GMS enrollment	16.4 (4.5) ¹	20.8 (3.9)
Years of culture data	16.0 (4.4) ¹	18.2 (3.8)
Age of transition ³ from OP to sputum cultures	7.6 (4.0) ¹	11.7 (5.6)
Respiratory culture data		
Age (years) first <i>Pa</i> -positive culture	5.8 (4.6) ¹	10.8 (5.7)
Age (years) persistent <i>Pa</i> -positive cultures	6.6 (4.7) ¹	12.3 (5.5)
Years from first culture data to first <i>Pa</i> -positive culture	2.8 (3.4) ¹	5.6 (5.0)
Never <i>Pa</i> positive by “first culture” definition	8 (3%)	17 (5%)
Never <i>Pa</i> positive by “persistent” definition	25 (8%) ¹	48 (15%)

¹ $P < 0.0001$ (Student's *t*-test).

² $P < 0.05$ (Student's *t*-test).

³ Transition age was defined as the age at the time of the first of three consecutive years with 1 sputum culture reported in at least 2 of the 3 years.

TABLE 2

Association Between Age of *Pseudomonas aeruginosa* Infection and Severity of Lung Disease

	Odds ratio (95% CI) for severe lung disease	
	Crude	Adjusted ¹
Age of first <i>Pseudomonas aeruginosa</i>		
0–4 years	9.6 (6.1, 15.0) ²	5.9 (3.2, 10.9) ²
5–9 years	3.2 (2.2, 4.8) ²	3.5 (2.1, 5.8) ²
10 years or older (or never <i>Pa</i> positive)	1 (referent)	1 (referent)
Age of persistent <i>Pseudomonas aeruginosa</i>		
0–4 years	18.6 (10.3, 33.9) ²	11 (5.5, 22.1) ²
5–9 years	4.3 (3.0, 6.4) ²	4.3 (2.7, 6.8) ²
10 years or older (or never persistent <i>Pa</i>)	1 (referent)	1 (referent)

¹ Adjusted for years of culture data, age of first culture data, CF center, and type of health insurance.

² $P < 0.0001$, compared to referent group.

TABLE 3

Association Between Age of *Pseudomonas aeruginosa* Infection and Severity of Lung Disease by Gender

	<u>Adjusted* odds ratio (95% CI) for severe lung disease</u>	
	Females (n = 301)	Males (n = 328)
Age of first <i>Pseudomonas aeruginosa</i>		
0–4 years	9.6 (4.1, 22) ^{1,2} (n = 91)	4.0 (1.8, 8.8) ¹ (n = 88)
5–9 years	7.8 (3.7, 16.3) ^{1,3} (n = 99)	1.8 (0.9, 3.4) (n = 105)
10 years or older (or never <i>Pa</i> positive)	1 (referent)	1 (referent)
Age of persistent <i>Pseudomonas aeruginosa</i>		
0–4 years	39.7 (12, 131.8) ^{1,3} (n = 54)	4.4 (1.9, 10.3) ¹ (n = 65)
5–9 years	11.7 (5.9, 23.5) ^{1,3} (n = 100)	1.7 (0.9, 3.3) (n = 96)
10 years or older (or never <i>Pa</i> positive)	1 (referent)	1 (referent)

* Adjusted for years of culture data, age of first culture data, CF center, and type of health insurance.

¹P = 0.001, compared to referent group.

²P = 0.1, comparing females to males by age group.

³P = 0.003, comparing females to males by age group.