Antibiotic Management of Lung Infections in Cystic Fibrosis

II. Nontuberculous Mycobacteria, Anaerobic Bacteria, and Fungi

James F. Chmiel[1], Timothy R. Aksamit[2], Sanjay H. Chotirmall[3], Elliott C. Dasenbrook[4], J. Stuart Elborn[5], John J. LiPuma[6], Sarath C. Ranganathan[7], Valerie J. Waters[8], and Felix A. Ratjen[9]

[1]Department of Pediatrics, Case Western Reserve University School of Medicine and Rainbow Babies and Children's Hospital, Cleveland, Ohio; [2]Division of Pulmonary Disease and Critical Care Medicine, Department of Medicine, Mayo Clinic, Rochester, Minnesota; [3]Department of Respiratory Medicine, Beaumont Hospital, Dublin, Republic of Ireland and Lee Kong Chian School of Medicine, Nanyang Technological University, Singapore; [4]Departments of Medicine and Pediatrics, Case Western Reserve University School of Medicine and University Hospitals Case Medical Center, Rainbow Babies and Children's Hospital, Cleveland, Ohio; [5]Medicine and Surgery, Queens University Belfast and Belfast City Hospital, Belfast, Northern Ireland, United Kingdom; [6]Department of Pediatrics and Communicable Diseases, University of Michigan Medical School, Ann Arbor, Michigan; [7]Department of Respiratory Medicine, Royal Children's Hospital, Infection and Immunity Theme, Murdoch Children's Research Institute, and Department of Pediatrics, University of Melbourne, Melbourne, Australia; and [8]Division of Infectious Diseases and [9]Division of Respiratory Medicine, Department of Pediatrics, University of Toronto and Hospital for Sick Children, Toronto, Ontario, Canada

Abstract

Airway infections are a key component of cystic fibrosis (CF) lung disease. Whereas the approach to common pathogens such as *Pseudomonas aeruginosa* is guided by a significant body of evidence, other infections often pose a considerable challenge to treating physicians. In Part I of this series on the antibiotic management of difficult lung infections, we discussed bacterial organisms including methicillin-resistant *Staphylococcus aureus*, gram-negative bacterial infections, and treatment of multiple bacterial pathogens. Here, we summarize the approach to infections with nontuberculous mycobacteria, anaerobic bacteria, and fungi. Nontuberculous mycobacteria can significantly impact the course of lung disease in patients with CF, but differentiation between colonization and infection is difficult clinically as coinfection with other micro-organisms is common. Treatment consists of different classes of antibiotics, varies in intensity, and is best guided by a team of specialized clinicians and microbiologists. The ability of anaerobic bacteria to contribute to CF lung disease is less clear, even though clinical relevance has been reported in individual patients. Anaerobes detected in CF sputum are often resistant to multiple drugs, and treatment has not yet been shown to positively affect patient outcome. Fungi have gained significant interest as potential CF pathogens. Although the role of *Candida* is largely unclear, there is mounting evidence that *Scedosporium* species and *Aspergillus fumigatus*, beyond the classical presentation of allergic bronchopulmonary aspergillosis, can be relevant in patients with CF and treatment should be considered. At present, however there remains limited information on how best to select patients who could benefit from antifungal therapy.

Keywords: anaerobic bacteria; *Aspergillus fumigatus*; *Mycobacterium abscessus*; *Mycobacterium avium* complex; *Scedosporium* species complex

(Received in original form May 12, 2014; accepted in final form August 7, 2014)

Supported by National Institutes of Health grant P30-DK27651. The U.S. Cystic Fibrosis Foundation is gratefully acknowledged.

Author Contributions: J.F.C. organized the manuscript; wrote the abstract, introduction, and summary; and edited the manuscript. E.C.D. reviewed, edited, and revised the manuscript and organized the references. J.J.L. reviewed, edited, and revised the manuscript. V.J.W. created Table 1 and reviewed, edited, and revised the manuscript. J.S.E. reviewed, edited, and revised the manuscript. T.R.A. was the lead author for the section on nontuberculous mycobacteria and created Figure 1. S.H.C. was the lead author for the section on fungi. S.C.R. was the lead author for the section on anaerobic bacteria. F.A.R. co-wrote the abstract, introduction, and summary, and served as the deciding editor of the manuscript.

Correspondence and requests for reprints should be addressed to James F. Chmiel, M.D., M.P.H., Division of Pediatric Pulmonology and Allergy/Immunology, Room 3001, Rainbow Babies and Children's Hospital, 11100 Euclid Avenue, Cleveland, OH 44106. E-Mail: james.chmiel@uhhospitals.org

Ann Am Thorac Soc Vol 11, No 8, pp 1298–1306, Oct 2014

DOI: 10.1513/AnnalsATS.201405-203AS
Internet address: www.atsjournals.org

Lung disease accounts for the majority of the morbidity and mortality in cystic fibrosis (CF) (1). Bacteria typically found in airway secretions of patients with CF include *Staphylococcus aureus*, *Haemophilus influenzae*, *Pseudomonas aeruginosa*, *Burkholderia cepacia* complex, *Stenotrophomonas maltophilia*, and *Achromobacter* species (2). Many of these bacteria have been associated with a decline in lung function in CF (3–6). However, other microorganisms have been isolated

from CF lung fluids including nontuberculous mycobacteria (NTM), anaerobic bacteria, and fungi. Although the isolation of some of these microorganisms is temporally associated with deterioration in baseline health in some individuals, it is not clear whether they are relevant pathogens in all patients. Clinicians must discern how to proceed if one of these organisms is isolated in the absence of clinical manifestations, changes in lung function, or radiographic changes. In addition, the clinician must also balance the potential for development of toxicities from prescribed therapies with the possibility of beneficial effect. Further complicating the decision on whether treatment should be initiated is the need to determine the best therapeutic regimen for the individual patient. These regimens may carry a large treatment burden and interfere with other therapies.

A scientific symposium was held at the 2013 American Thoracic Society (ATS) International Conference in Philadelphia, Pennsylvania to discuss the management of difficult-to-treat lung infections in CF. From this symposium, two companion manuscripts were written to summarize the current evidence presented at that meeting. In Part I, the lung microbiome, methicillin-resistant *S. aureus*, gram-negative bacteria, and the treatment of multiple infections in CF were discussed (7). In this article (Part II), NTM, anaerobic bacteria, and fungi are discussed. Practical treatment approaches are discussed in both articles. These approaches summarize the current evidence. However, it is imperative to recognize that these treatment approaches are evolving as the results of ongoing and future research studies become available. The reader should realize that these documents are not portrayed as guideline documents or consensus recommendations. The discussions contained within these articles are not meant to represent the finish line for treating lung infections in CF but rather should be taken to represent the starting line.

Nontuberculous Mycobacteria

NTM lung infections are increasingly observed in the general population and in patients with CF (8). The incidence of NTM lung disease in patients with CF, which most often occurs in patients older than 15 years and increases with age, has been estimated to be 13–20% (9–11). *Mycobacterium avium* complex (MAC) and *Mycobacterium abscessus* represent the two most common NTM species causing infection in CF. Accelerated loss of lung function has been observed in patients with *M. abscessus* (12). In patients with CF under evaluation for lung transplantation, up to 20% of patients were found to have NTM (13). *M. abscessus* in particular is associated with worse outcome and the need for NTM treatment posttransplantation (13). As a result, many transplant centers now consider the presence of NTM lung disease a relative contraindication for transplantation (13).

NTM is ubiquitous in water and soil, and can frequently be isolated from residential sources including showerheads and other home water sources (14–16). Peat moss exposure in some studies has been identified as a potential exposure risk for NTM lung disease (14, 17, 18). However, case–control studies have not clearly demonstrated an association between exposure to residential water sources or other activities including gardening and NTM lung disease (19). Furthermore, patient NTM isolates do not always match environmental NTM isolates, matching in 22–41% of cases when the same MAC species were isolated (14, 16). Nonetheless, certain environmental precautions may reduce exposure. Simple environmental controls implemented at home that may reduce exposure to NTM include increasing the temperature of hot water heaters to greater than 130°F, installing large droplet showerheads to reduce aerosolization, avoiding tap water rinses of equipment and avoiding tap water mouth rinses before sputum collection, and avoiding peat moss dust exposure by wearing a facemask and moistening the soil before working with it (18, 20). It is not clear, however, whether these risk modifications impact the development of NTM lung disease. Reports of the transmissibility of *M. abscessus* species in two outpatient CF clinics is worth noting given the previous experience of lack of human-to-human transmissibility of NTM lung disease (21, 22). Although both reports describe the transmissibility of *M. abscessus* ssp. *massiliense*, it is unclear whether the risk of transmission is restricted to only this subspecies or whether it may be generalized to all *M. abscessus* or even other NTM isolates. Therefore, close collaboration with the infection control team and abiding by infection control procedures in CF and bronchiectasis clinics, including respiratory isolation for patients with *M. abscessus*, is warranted in outpatient as well as inpatient settings to prevent transmission of *M. abscessus* in high-risk situations.

The diagnosis of NTM lung disease is based on criteria outlined by the ATS, including a combination of clinical, radiographic, and microbiologic elements (9). It is worth noting that in most circumstances and for most NTM respiratory isolates (especially MAC), one positive culture, especially with low numbers of organisms, smear negative, or growth on liquid media only, is not adequate to establish a diagnosis of NTM lung disease. A presumptive diagnosis based on clinical and radiographic features is equally inappropriate for the initiation of empiric therapy. As such, longitudinal monitoring and multiple cultures may be required before a diagnosis of NTM lung disease is firmly established. Further contributing to this conundrum is the well-recognized waxing and waning of radiographic abnormalities, without overall radiographic progression, that is known to occur in patients with NTM lung disease regardless of treatment status. Establishing a more certain diagnosis before committing to treatment of NTM lung disease is thus required. Overdiagnosis results in the unnecessary exposure to complicated drug regimens, and underdiagnosis may result in the development of progressive lung disease with irreversible loss of lung function from inadequate treatment.

The decision regarding whether to treat NTM lung disease must therefore balance the risks and benefits of treatment versus observation. Individualized patient factors include the risk of progression, goals of therapy (sputum conversion vs. suppression), status of comorbid medical conditions (gastroesophageal reflux, sinus disease, and bronchiectasis), medication tolerance, and patient acceptance (Figure 1). Similarly, the intensity of the NTM regimen should be proportionate to disease severity and goals of therapy. In the case of MAC lung disease, the treatment options range from observation to three times weekly oral therapy to daily therapy plus parenteral therapy. Special attention regarding

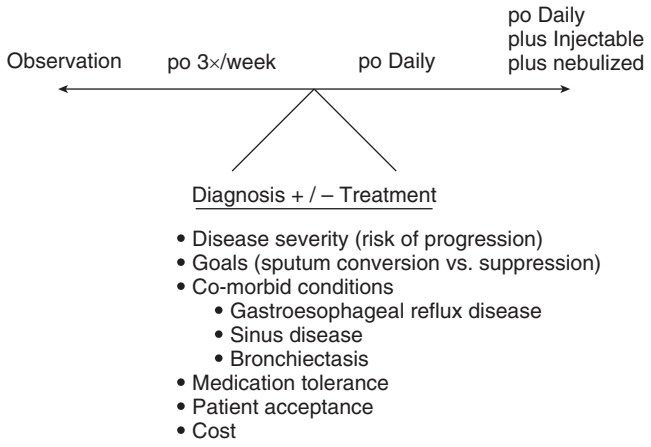

Figure 1. Determining treatment of nontuberculous mycobacteria (NTM) lung disease requires assessment of diagnostic strategies, treatment options, and individualized risk–benefit analyses. When deciding whether to treat a patient with NTM lung disease, the risk and benefits of treatment must be weighed against observation. This decision is influenced by many factors including the risk of progression, goals of therapy, and patient factors. Po = per os (by mouth).

macrolide use for antiinflammatory purposes is warranted to avoid the development of resistance in macrolide-susceptible NTM lung disease (23–27). CF guidelines have reaffirmed the need to screen patients at baseline and then every 6–12 months (28). This recommendation has not changed despite preliminary data not finding increased macrolide resistance in CF patients with NTM infections receiving chronic macrolide therapy (29).

Specific treatment regimens for NTM lung disease are outlined in the ATS statement (9). Although these recommendations remain appropriate for treating MAC lung disease, the use of nebulized amikacin and increased understanding of the various treatment approaches for *M. abscessus* lung disease warrant special comment. Inhaled amikacin has been increasingly used for the treatment of both MAC and *M. abscessus* lung disease despite limited published clinical experience (30–32). Dosing of nebulized amikacin is variable but most often ranges between 250 and 500 mg once or twice daily. Higher dosing is generally less well tolerated (32). Results from a completed phase 2 study of liposomal amikacin for refractory NTM lung disease are expected to be released soon. The use of nebulized amikacin when treating NTM lung disease should be considered as part of a multidrug mycobacterial regimen.

The recognition of a variably expressed erythromycin ribosomal methylation (*erm*) gene in *M. abscessus* has been associated with variable clinical response to macrolide-based regimens for *M. abscessus* (33–35). This may also explain some of the apparent macrolide resistance in other NTM organisms such as *M. fortuitum*. This is in contrast to *M. chelonae*, which does not express an active *erm* gene. The *erm* gene encodes enzymes that methylate the 23S ribosomal RNA within the 50S ribosomal subunit, resulting in reduced binding affinity of macrolides for their specific target, to impair protein synthesis. The additional importance of the presence or absence of an active *erm* gene is highlighted by the difference between *M. abscessus* subspecies. *M. abscessus* ssp. *abscessus* has universal expression of an active *erm* gene conferring macrolide resistance whereas other *M. abscessus* subspecies, such as *M. abscessus* ssp. *bolletii*, also known as *M. massiliense*, do not express an active *erm* gene. It is important to note, however, that even in the absence of the *erm* gene, NTM species may be resistant to macrolides through other mechanisms. Expression of the *erm* gene can be variable; there are some data to suggest that clarithromycin induces greater expression of the gene than does azithromycin (34). The inclusion of a macrolide in treatment regimens for *M. abscessus* lung disease thus relies heavily on the presence or absence of an active *erm* gene or, as a surrogate, differentiation of *M. abscessus* ssp. *abscessus* from *M. abscessus* ssp. *bolletii* (*M. massiliense*). Initial *M. abscessus* isolates should be incubated with macrolide for 14 days before a determination of macrolide susceptibility and, by inference, the presence or absence of an active *erm* gene. Other potential mechanisms of drug resistance have been described, but the clinical significance of these requires further study (36). Thus, the approach to the treatment of *M. abscessus* ssp. *abscessus* lung disease most often involves a regimen including other nonmacrolide agents, such as amikacin with a combination of two or more additional antibiotics including cefoxitin or imipenem, tigecycline, or linezolid. In the absence of inducible macrolide resistance or an active *erm* gene, regimens for *M. abscessus* ssp. *bolletii* (*M. massiliense*) should include clarithromycin or azithromycin in addition to multiple other antibiotics. A typical practice pattern, even without clear supporting data in the literature, is to begin with an intensive treatment regimen including both parenteral and oral agents followed by deescalation to an inhaled and oral regimen after a period of weeks or months. The timing and specifics of this transition can be particularly variable given the essential need to avoid monotherapy and to maintain a multidrug regimen with effective nonparenteral agents; the efficacy of which must be weighed against the risks of toxicity and the technical challenges of extended use of parenteral agents. Consultation with a pulmonary disease, infectious disease, and/or NTM expert is generally recommended. Common treatment regimens for NTM lung disease are given in Table 1. Surgical resection in conjunction with medical therapy should be considered for localized cavitary NTM lung disease, macrolide-resistant MAC lung disease, and *M. abscessus* ssp. *abscessus* lung disease in highly selected patients. Surgery, when considered, should be undertaken by an experienced team of mycobacterial physicians including surgeons with robust experience in mycobacterial lung surgery.

In summary, NTM lung disease in patients with CF presents variably and remains a complex problem with respect to establishing a diagnosis and treatment program when indicated. Longitudinal follow-up may be required before specific treatment recommendations can be made. Patients with CF with NTM lung disease are best cared for by teams of clinicians experienced in the care of patients with mycobacterial infections and who work closely with their laboratory colleagues to optimize the timing and intensity of multidrug mycobacterial lung disease

Table 1. Empiric antibiotic therapy for the treatment of nontuberculous mycobacteria lung infections in cystic fibrosis*

Organism	Antibiotic	Pediatric Dose	Adult Dose	Side Effects
Mycobacterium abscessus	Clarithromycin or azithromycin†	Clarithromycin 15 mg/kg orally (max 500 mg) twice daily or azithromycin 5 mg/kg/d (max 250 mg)	Clarithromycin 500 mg orally twice daily or azithromycin 250–500 mg orally daily	GI, ototoxicity
	Plus amikacin‡	10–30 mg/kg intravenously daily or 25–30 mg/kg three times weekly followed by 250–500 mg nebulized daily to twice daily	10–30 mg/kg intravenously daily or 25–30 mg/kg three times weekly followed by 250–500 mg nebulized daily to twice daily	Ototoxicity, nephrotoxicity
	And cefoxitin	200–250 mg/kg/d in divided doses (max 12 g)	200–250 mg/kg/d in divided doses (max 12 g)	GI, rash, myelosuppression
	Or imipenem	60–100 mg/kg/d intravenously divided doses (max 2 g)	1–2 g intravenously divided doses	GI, rash, myelosuppression, rarely seizures
	Or tigecycline	1.2 mg/kg intravenously every 12 h (max 50 mg)	25–50 mg daily intravenously	GI, cholestasis, myelosuppression
	Or linezolid (include pyridoxine 50 mg daily)	If < 11 yr: 10 mg/kg intravenously or orally every 8 h If > 11 yr: 10 mg/kg (max 600 mg) intravenously or orally daily to twice daily	300–600 mg intravenously or orally daily to twice daily	Optic/peripheral neuropathy, myelosuppression
Mycobacterium avium complex	Clarithromycin or azithromycin	Clarithromycin 15 mg/kg orally (max 500 mg) twice daily or azithromycin 5 mg/kg/d (max 250 mg)	Clarithromycin 500 mg orally twice daily or azithromycin 250–500 mg orally daily	GI, ototoxicity
	Plus rifampin	10–20 mg/kg orally once daily (max 600 mg)	450–600 mg orally once daily	Hepatotoxicity, body fluid discoloration
	And ethambutol	15 mg/kg orally once daily	15 mg/kg orally once daily	Optic/peripheral neuritis
	Plus for *advanced disease*: amikacin‡	10–30 mg/kg intravenously daily or 25–30 mg/kg three times weekly followed by 250–500 mg nebulized daily to twice daily	10–30 mg/kg intravenously daily or 25–30 mg/kg three times weekly followed by 250–500 mg nebulized daily to twice daily	Ototoxicity, nephrotoxicity

Definition of abbreviations: GI = gastrointestinal; min, minimum; max, maximum.
*The antibiotic doses given come from a compilation of sources and practice patterns including commonly prescribed off-label doses and uses. Sources include the pharmacy formulary of the Hospital for Sick Children (Toronto, ON, Canada), which is based on product inserts and the published literature. The doses given are general guidelines, and may vary somewhat between institutions. It is recommended that the clinician consult his/her institution's pharmacy, product inserts, and published literature before prescribing these drugs. Consultation with a pulmonary disease, infectious disease, and/or nontuberculous mycobacteria expert to individualize treatment regimens is recommended.
†Consider alternative antibiotic if the *erm* gene (encoding inducible macrolide resistance) is detected or inducible macrolide resistance is noted.
‡Serum concentrations should be monitored, and aim for a maximal serum concentration (Cmax) in the range of 80–120 mg/L with a minimal serum concentration (Cmin) of less than 1 mg/L. Alternatively, peak levels may also be used with a target peak serum level between 20 and 35 μg/ml. It is known that patients with CF generally have an increased volume of distribution and more rapid clearance, which may require higher dosing than for others without CF.

treatment regimens, carefully weighing risks and benefits and, when necessary, also considering surgical intervention.

Anaerobic Bacteria

Anaerobes are organisms that do not require oxygen for growth. They can be obligate or facultative; *P. aeruginosa* is an example of the latter. In this state, *P. aeruginosa* exists as a slow-growing organism that is relatively resistant to antibiotics.

Obligate anaerobes have been implicated in a number of non-CF infections, such as infections of the upper respiratory tract and aspiration pneumonia. Steep oxygen gradients exist within CF mucus such that even at relatively shallow depths within mucus, the environment is considered to be hypoxic, or even frankly anaerobic (37). Conventional culture-dependent approaches are not optimized for identifying anaerobes. Specific anaerobic culture methods, or culture-independent techniques, may be more appropriate. Several studies on tracheal aspirates, sputum, or bronchoalveolar lavage (BAL) fluid have confirmed the presence of anaerobes in the lower airways in CF in up to 80% samples and at bacterial densities of between 10^7 and 10^9 colony-forming units/ml in sputum (38–43). The most common genera identified were *Prevotella*, *Veillonella*, *Propionibacterium*, *Actinomyces*, *Staphylococcus saccharolyticus*, *Peptostreptococcus*, and *Clostridium* (44). Using terminal restriction fragment

length polymorphism, Rogers and colleagues (45) identified differences between paired mouthwash and sputum samples obtained from subjects with CF, both in the bands identified and the band volume, suggesting that the finding of anaerobes in the lower airways is not explained by aspiration of the oral anaerobiota.

Although the inflammatory response to individual aerobic organisms identified in BAL fluid from infants and young children with CF has been described (46), no such data linking anaerobes to inflammation or clinical outcomes are available. Studies in younger subjects might help to elucidate the role of anaerobes in disease pathogenesis and whether or not their presence in the lower airways represents an epiphenomenon (47). Existing studies are in older subjects who have therefore experienced a more complex infection history. Longitudinal studies are also lacking, although a comprehensive longitudinal study conducted in a single patient suggested that anaerobes of the *Streptococcus milleri* group contributed to the development of pulmonary exacerbations (48). Ulrich and colleagues reported that 16 of 17 patients with CF produced antibodies against two immunoreactive antigens of *Prevotella intermedia* compared with 0 of 30 controls (49), suggesting that anaerobes are, indeed, immunogenic in CF. Culture supernatant fluid of *P. intermedia* was also cytotoxic to respiratory epithelial cell lines, associated with neutrophil and macrophage recruitment into lung tissue in mice, and cytotoxic to human-derived neutrophils. Its pathogenicity is estimated as being intermediate between that of aerobic and anaerobic *P. aeruginosa*. In studies where anaerobes were specifically targeted during treatment for pulmonary exacerbations, the results have been conflicting. Worlitzsch and colleagues (43) did not identify any significant reduction in the density of anaerobes in sputum after treatment with antibiotics despite an increase in pulmonary function during the period of treatment. Similarly, Tunney and colleagues identified only limited reduction in the density of anaerobes at the end of 2 weeks of treatment (50). An important factor to consider when treating anaerobic infections is that anaerobic organisms are often resistant to the commonly administered antibiotics. For example, resistance to metronidazole was reported to occur in nearly all *Peptostreptococcus* and *Streptococcus* species, whereas resistance to meropenem is more rare (44). Although meropenem is commonly included in CF antibiotic protocols as a second- or third-line intravenous drug in the treatment of pulmonary exacerbations, it remains unclear whether any clinical improvements associated with its administration are related directly to its targeting of anaerobes.

Data from culture-based studies, and more recently from studies using culture-independent techniques, therefore indicate that anaerobes are prevalent in the lower airways of people with CF, but whether these organisms play a part in the pathophysiology of progressive lung damage remains unknown. How anaerobes interact with the microbiota of the lower airways and other CF organisms also requires study. Resistance *in vitro* is common, meaning that antibiotics usually considered for the treatment of anaerobes may not be effective. Anaerobes appear to play a role in CF lung disease, but this requires clarification before the targeting of obligate anaerobes in the treatment of CF lung infections becomes routine.

Fungi

Patients with CF are at increased risks of fungal colonization owing to impaired mucus clearance, local immunogenic dysfunction, and antibiotic use. Although CF lung disease is classically dominated by bacteria, fungal isolates are increasingly described because the respiratory tract anatomically communicates with the atmosphere, a rich source of airborne fungal spores. Inability to clear such inhaled particles results in their persistence, colonization, and potential airway infection. This spectrum of clinical consequences combined with enhanced detection methods makes it probable that we have thus far underestimated fungal prevalence and importance in clinical practice over the last decade of CF care (51–58).

Vast arrays of fungal species are described in CF; however, methods used for their isolation primarily dictate the species and populations detected. Although traditional methodologies of fungal culture remain, emerging molecular techniques and genotyping provide greater sensitivity. Despite fungal biodiversity (Figure 2), the major clinical challenges are caused by *Aspergillus fumigatus* (59, 60), *Candida albicans* (61, 62), and *Scedosporium* species complex. Clinicians are often left wondering about the significance of isolating fungi from a patient with CF and whether treatment is indicated.

A. fumigatus is detected in sputum in approximately 30% of patients with CF. Allergic bronchopulmonary aspergillosis (ABPA) remains a key consequence, but sputum isolation does not correlate with ABPA occurrence (63). The difficulty in

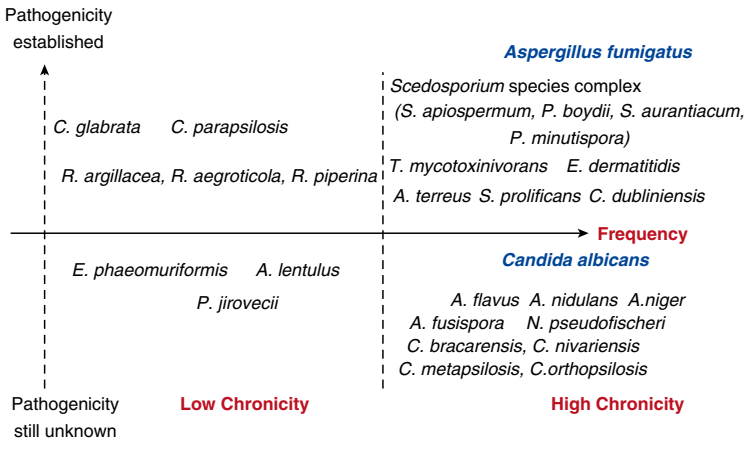

Figure 2. Cystic fibrosis fungal biodiversity grouped according to frequency of isolation (*x* axis) and established pathogenicity (*y* axis). The fungi are further divided in terms of chronicity as illustrated. The most frequently isolated filamentous fungi, *Aspergillus fumigatus* and *Scedosporium* species complex, and yeast *Candida albicans* are highlighted and further discussed in text. Low-chronicity genera: *A.* = *Aspergillus*; *C.* = *Candida*; *E.* = *Exophiala*; *P.* = *Pneumocystis*; *R.* = *Rasamsonia.* High-chronicity genera: *A.* = *Aspergillus* (*flavus, nidulans, niger*); *A.* = *Acrophialophora* (*fusispora*); *C.* = *Candida*; *E.* = *Exophiala*; *N.* = *Neosartorya*; *P.* = *Pseudallescheria*; *S.* = *Scedosporium*; *T.* = *Trichosporon*.

diagnosis of ABPA exists because of overlapping clinical, radiological, immunological, and microbiological features similar to that of an infective exacerbation in CF (63, 64). To address this, biomarkers such as recombinant *Aspergillus* antigens, precipitins, anti-*Aspergillus* IgG, thymus activation and regulated chemokine (TARC), and the basophil surface marker CD203c have been proposed, but pose difficulties because of their variability and lack of sensitivity, standardization, and accessibility (65). In acute ABPA, corticosteroids suppress the inflammatory response. One treatment protocol employed includes prednisone at 40 mg once daily for 2 weeks with taper over 3 months tailored to clinical symptoms, lung function, and total serum IgE concentration. Concerns over side effects of long-term steroid administration has prompted the use of alternative regimens such as high-dose methylprednisolone (10–15 mg/kg) daily for 3 days monthly for up to 10 months (66). Antifungal therapy may be concurrently administered. However, no randomized controlled trials to date support use in CF-ABPA (67). Itraconazole is preferred with a favorable side effect profile, but it does possess variable absorption and food interactions necessitating close serum monitoring. In addition, the development of azole resistance remains a concern (68). Voriconazole is an alternative drug option but has significant associated photosensitivity especially in patients with CF (69). Steroid-resistant cases may necessitate antifungal therapy or administration of an anti-IgE monoclonal antibody, but existing evidence is limited to case series reports (70).

Clinically distinct from ABPA, *Aspergillus* sensitization independently affects pulmonary function; however, the mechanism through which it does so remains unclear (71). Allergic sensitization does not correlate with sputum detection of *Aspergillus*. Unlike *Candida* sensitization, *Aspergillus* sensitization is associated with greater lung function decline and pulmonary exacerbations (72). The presence of severe CF mutations, mild lung disease ($FEV_1 > 70\%$), absence of *Pseudomonas*, and prior azithromycin exposure all remain predictive for *Aspergillus* sensitization (73). A novel immunological classification for CF aspergillosis has been proposed. On the basis of serum IgE and IgG concentrations combined with sputum galactomannan and the presence of PCR-detectable *Aspergillus*, four distinct subgroups are defined. These include those with ABPA, those who are *Aspergillus* sensitized, those with *Aspergillus* bronchitis, and those without disease. Improved classification and definition can assist with clinical phenotyping and may impact future treatment decisions in *Aspergillus*-associated CF disease (74).

Controversy persists over the significance of non-ABPA *Aspergillus* colonization. It is often associated with worse radiologic findings and is an independent risk factor for hospitalization (75, 76). Itraconazole treatment reduces the burden of *Aspergillus*, attenuates radiological mosaic perfusion, reduces exacerbations, and stabilizes pulmonary function in this setting (77). Such effects are mediated by down-regulation of the vitamin D receptor through the virulence factor gliotoxin. Itraconazole treatment has been shown to decrease BAL gliotoxin concentrations and to restore vitamin D receptor expression with concomitant reduction in helper T-cell type 2 cytokines IL-5 and IL-13, drivers of ABPA (77). Despite these findings, a double-blind, placebo-controlled trial failed to demonstrate clinical benefit, but treatment efficacy may have been impacted by failing to achieve therapeutic itraconazole concentrations in a significant proportion of patients (78). Further study in this area is warranted before treatment recommendations can be issued (if necessary) for the non-ABPA *Aspergillus*–colonized population.

C. albicans is capable of causing oral and genital candidiasis and vascular device infections in CF (62, 79). It is frequently isolated from CF sputum. Patients with CF are at increased risk of pulmonary colonization because of inhaled steroid use, CF-related diabetes, and lifelong antibiotic exposure. A prospective longitudinal study showed high (49.4%) colonization rates best predicted by pancreatic insufficiency, osteopenia, and cocolonization with *P. aeruginosa*, all features of advanced disease (61). Colonization presaged increases in hospitalizations for exacerbations and longitudinal declines in body mass index and FEV_1 (61). At present, its clinical role (if any) is unclear, and there is no evidence to suggest treatment benefit.

Members of the *Scedosporium* species complex are chronic colonizers and emerging pathogens in CF (80, 81). A major risk exposure includes potted plants. However, they also have an environmental presence (82, 83). Colonization is not associated with FEV_1 or steroid or antifungal use. Interestingly, those harboring the fungus are less likely to be colonized with *P. aeruginosa* (84). Discordance between relatively high isolation frequency (6.5–10%) and low environmental abundance prompts questions about how initial acquisition actually occurs in CF (80, 81). Genotype analysis of sequential isolates demonstrates that individual patients are colonized by unique phenotypes that remain conserved over time (85). Clinical consequences include allergic responses and risk of dissemination in immunocompromised hosts (86). Eradication remains difficult once colonization is established, with voriconazole the agent of choice.

Although our knowledge regarding the role of fungi in CF is improving, many questions remain. Are certain fungi pathogenic and if so, what mechanisms do they use? When do they become pathogenic? Are they pathogenic from the time they enter into the airway or only after a certain time of colonization and sensitization? Does clinical setting matter? Should attempts be made to eradicate them? If so when, with what drugs, and for how long? These are all valid questions, which are difficult to answer on the basis of existing data (60).

There is limited knowledge regarding treatment approaches for fungi in CF. *A. fumigatus* is commonly detected in the CF airway. It is a proven fungal pathogen in CF-ABPA. Sputum isolation is discordant with ABPA occurrence, thereby making diagnosis difficult. Treatment should always be pursued in CF-ABPA. However, it remains controversial in the non-ABPA *Aspergillus*–colonized patient. There is no evidence that *C. albicans* isolated from CF sputum should be treated because its pathological significance in the airway is unknown. No current evidence exists to suggest treatment benefit in this context. However, when *C. albicans* causes mucosal or vascular device infection, prompt treatment is indicated. Infection rates with *Scedosporium* species are underestimated

because of difficulties with diagnosis, as this mold's clinical, radiological, and pathological appearance is similar to *Aspergillus*. Such misdiagnosis may be lethal considering that *Scedosporium* is almost always resistant to amphotericin B, the agent frequently used in presumptive *Aspergillus* infection. Consequently, eradication should be attempted at first isolation in view of its potentially devastating clinical consequences if misdiagnosed or allowed to persist long term.

Summary

For decades, clinicians have been treating a narrow array of bacteria that infect the CF airway (2). Under selective pressure of frequent antibiotic use and with improved techniques to identify microorganisms, that array is expanding. Physicians must treat not only the classic pathogens associated with CF, such as *S. aureus* and *P. aeruginosa*, they may also have to treat other microorganisms such as NTM, anaerobic bacteria, and fungi. Less evidence regarding treatment of these organisms is available than for the typical bacteria known to infect the CF airway. These organisms often grow slowly, if at all, on typical microbiological cultures. However, when a new organism is identified, CF clinicians are often left wondering about the pathologic significance of this new finding. Furthermore, determining a treatment regimen is often frustrating to even the most experienced individual. The airway environment in CF is continually evolving. Niches are being created that will allow new potential pathogens to gain a foothold in the CF airway. Therefore, clinicians must be constantly vigilant for the emergence of new microorganisms infecting the CF airway, and researchers must be prepared to develop novel antimicrobial therapies to treat these infections. ■

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

Acknowledgment: The authors acknowledge Jay Hilliard for help with editing and refining the figures and tables.

References

1 Davis PB. Cystic fibrosis since 1938. *Am J Respir Crit Care Med* 2006; 173:475–482.
2 Chmiel JF, Davis PB. State of the art: why do the lungs of patients with cystic fibrosis become infected and why can't they clear the infection? *Respir Res* 2003;4:8.
3 Courtney JM, Dunbar KE, McDowell A, Moore JE, Warke TJ, Stevenson M, Elborn JS. Clinical outcome of *Burkholderia cepacia* complex infection in cystic fibrosis adults. *J Cyst Fibros* 2004; 3:93–98.
4 Dasenbrook EC, Merlo CA, Diener-West M, Lechtzin N, Boyle MP. Persistent methicillin-resistant *Staphylococcus aureus* and rate of FEV_1 decline in cystic fibrosis. *Am J Respir Crit Care Med* 2008;178: 814–821.
5 Demko CA, Byard PJ, Davis PB. Gender differences in cystic fibrosis: *Pseudomonas aeruginosa* infection. *J Clin Epidemiol* 1995;48: 1041–1049.
6 Kerem E, Viviani L, Zolin A, MacNeill S, Hatziagorou E, Ellemunter H, Drevinek P, Gulmans V, Krivec U, Olesen H; ECFS Patient Registry Steering Group. Factors associated with FEV_1 decline in cystic fibrosis: analysis of the ECFS Patient Registry. *Eur Respir J* 2014;43: 125–133.
7 Chmiel JF, Aksamit TR, Chotirmall SH, Dasenbrook EC, Elborn JS, LipPuma JJ, Ranganathan SC, Waters V, Ratjen F. Antibiotic management of lung infections in cystic fibrosis: the microbiome, MRSA, gram-negative bacteria, and multiple infections. *Ann Am Thorac Soc* 2014;11:1120–1129.
8 Kendall BA, Winthrop KL. Update on the epidemiology of pulmonary nontuberculous mycobacterial infections. *Semin Respir Crit Care Med* 2013;34:87–94.
9 Griffith DE, Aksamit T, Brown-Elliott BA, Catanzaro A, Daley C, Gordin F, Holland SM, Horsburgh R, Huitt G, Iademarco MF, *et al*.; ATS Mycobacterial Diseases Subcommittee; American Thoracic Society; Infectious Diseases Society of America. An official ATS/IDSA statement: diagnosis, treatment, and prevention of nontuberculous mycobacterial diseases. *Am J Respir Crit Care Med* 2007;175: 367–416. [Published erratum appears in *Am J Respir Crit Care Med* 175:744–745.]
10 Olivier KN, Weber DJ, Wallace RJ Jr, Faiz AR, Lee JH, Zhang Y, Brown-Elliot BA, Handler A, Wilson RW, Schechter MS, *et al*.; Nontuberculous Mycobacteria in Cystic Fibrosis Study Group. Nontuberculous mycobacteria. I. Multicenter prevalence study in cystic fibrosis. *Am J Respir Crit Care Med* 2003;167: 828–834.
11 Roux AL, Catherinot E, Ripoll F, Soismier N, Macheras E, Ravilly S, Bellis G, Vibet MA, Le Roux E, Lemonnier L, *et al*.; Jean-Louis Herrmann for the OMA Group. Multicenter study of prevalence of nontuberculous mycobacteria in patients with cystic fibrosis in France. *J Clin Microbiol* 2009;47:4124–4128.
12 Esther CR Jr, Esserman DA, Gilligan P, Kerr A, Noone PG. Chronic *Mycobacterium abscessus* infection and lung function decline in cystic fibrosis. *J Cyst Fibros* 2010;9:117–123.
13 Chalermskulrat W, Sood N, Neuringer IP, Hecker TM, Chang L, Rivera MP, Paradowski LJ, Aris RM. Non-tuberculous mycobacteria in end stage cystic fibrosis: implications for lung transplantation. *Thorax* 2006;61:507–513.
14 Falkinham JO III. Nontuberculous mycobacteria from household plumbing of patients with nontuberculous mycobacteria disease. *Emerg Infect Dis* 2011;17:419–424.
15 Feazel LM, Baumgartner LK, Peterson KL, Frank DN, Harris JK, Pace NR. Opportunistic pathogens enriched in showerhead biofilms. *Proc Natl Acad Sci USA* 2009;106:16393–16399.
16 Nishiuchi Y, Maekura R, Kitada S, Tamaru A, Taguri T, Kira Y, Hiraga T, Hirotani A, Yoshimura K, Miki M, *et al*. The recovery of *Mycobacterium avium-intracellulare* complex (MAC) from the residential bathrooms of patients with pulmonary MAC. *Clin Infect Dis* 2007;45:347–351.
17 De Groote MA, Pace NR, Fulton K, Falkinham JO III. Relationships between *Mycobacterium* isolates from patients with pulmonary mycobacterial infection and potting soils. *Appl Environ Microbiol* 2006;72:7602–7606.
18 Falkinham JO III. Reducing human exposure to *Mycobacterium avium*. *Ann Am Thorac Soc* 2013;10:378–382.
19 Kim RD, Greenberg DE, Ehrmantraut ME, Guide SV, Ding L, Shea Y, Brown MR, Chernick M, Steagall WK, Glasgow CG, *et al*. Pulmonary nontuberculous mycobacterial disease: prospective study of a distinct preexisting syndrome. *Am J Respir Crit Care Med* 2008; 178:1066–1074.
20 Saiman L, Siegel J; Cystic Fibrosis Foundation. Infection control recommendations for patients with cystic fibrosis: microbiology, important pathogens, and infection control practices to prevent patient-to-patient transmission. *Infect Control Hosp Epidemiol* 2003; 24(5, Suppl):S6–S52.
21 Aitken ML, Limaye A, Pottinger P, Whimbey E, Goss CH, Tonelli MR, Cangelosi GA, Dirac MA, Olivier KN, Brown-Elliott BA, *et al*. Respiratory outbreak of *Mycobacterium abscessus* subspecies *massiliense* in a lung transplant and cystic fibrosis center. *Am J Respir Crit Care Med* 2012;185:231–232.
22 Bryant JM, Grogono DM, Greaves D, Foweraker J, Roddick I, Inns T, Reacher M, Haworth CS, Curran MD, Harris SR, *et al*. Whole-genome sequencing to identify transmission of *Mycobacterium abscessus* between patients with cystic fibrosis: a retrospective cohort study. *Lancet* 2013;381:1551–1560.

23 Altenburg J, de Graaff CS, Stienstra Y, Sloos JH, van Haren EH, Koppers RJ, van der Werf TS, Boersma WG. Effect of azithromycin maintenance treatment on infectious exacerbations among patients with non-cystic fibrosis bronchiectasis: the BAT randomized controlled trial. *JAMA* 2013;309:1251–1259.

24 Ratjen F, Saiman L, Mayer-Hamblett N, Lands LC, Kloster M, Thompson V, Emmett P, Marshall B, Accurso F, Sagel S, *et al*. Effect of azithromycin on systemic markers of inflammation in patients with cystic fibrosis uninfected with *Pseudomonas aeruginosa*. *Chest* 2012;142:1259–1266.

25 Serisier DJ, Martin ML, McGuckin MA, Lourie R, Chen AC, Brain B, Biga S, Schlebusch S, Dash P, Bowler SD. Effect of long-term, low-dose erythromycin on pulmonary exacerbations among patients with non-cystic fibrosis bronchiectasis: the BLESS randomized controlled trial. *JAMA* 2013;309:1260–1267.

26 Southern KW, Barker PM, Solis-Moya A, Patel L. Macrolide antibiotics for cystic fibrosis. *Cochrane Database Syst Rev* [serial on the Internet]. 2012 [accessed September 2014];11:CD002203. Available from: http://summaries.cochrane.org/

27 Wong C, Jayaram L, Karalus N, Eaton T, Tong C, Hockey H, Milne D, Fergusson W, Tuffery C, Sexton P, *et al*. Azithromycin for prevention of exacerbations in non-cystic fibrosis bronchiectasis (EMBRACE): a randomised, double-blind, placebo-controlled trial. *Lancet* 2012; 380:660–667.

28 Mogayzel PJ Jr, Naureckas ET, Robinson KA, Mueller G, Hadjiliadis D, Hoag JB, Lubsch L, Hazle L, Sabadosa K, Marshall B; Pulmonary Clinical Practice Guidelines Committee. Cystic fibrosis pulmonary guidelines: chronic medications for maintenance of lung health. *Am J Respir Crit Care Med* 2013;187:680–689.

29 Binder AM, Adjemian J, Olivier KN, Prevots DR. Epidemiology of nontuberculous mycobacterial infections and associated chronic macrolide use among persons with cystic fibrosis. *Am J Respir Crit Care Med* 2013;188:807–812.

30 Davis KK, Kao PN, Jacobs SS, Ruoss SJ. Aerosolized amikacin for treatment of pulmonary *Mycobacterium avium* infections: an observational case series. *BMC Pulm Med* 2007;7:2.

31 Colin AA, Ali-Dinar T. Aerosolized amikacin and oral clarithromycin to eradicate *Mycobacterium abscessus* in a patient with cystic fibrosis: an 8-year follow-up. *Pediatr Pulmonol* 2010;45:626–627.

32 Leung JM, Olivier KN. Nontuberculous mycobacteria in patients with cystic fibrosis. *Semin Respir Crit Care Med* 2013;34:124–134.

33 Nash KA, Brown-Elliott BA, Wallace RJ Jr. A novel gene, *erm*(41), confers inducible macrolide resistance to clinical isolates of *Mycobacterium abscessus* but is absent from *Mycobacterium chelonae*. *Antimicrob Agents Chemother* 2009;53:1367–1376.

34 Choi GE, Shin SJ, Won CJ, Min KN, Oh T, Hahn MY, Lee K, Lee SH, Daley CL, Kim S, *et al*. Macrolide treatment for *Mycobacterium abscessus* and *Mycobacterium massiliense* infection and inducible resistance. *Am J Respir Crit Care Med* 2012;186:917–925.

35 Koh WJ, Jeon K, Lee NY, Kim BJ, Kook YH, Lee SH, Park YK, Kim CK, Shin SJ, Huitt GA, *et al*. Clinical significance of differentiation of *Mycobacterium massiliense* from *Mycobacterium abscessus*. *Am J Respir Crit Care Med* 2011;183:405–410.

36 van Ingen J, Boeree MJ, van Soolingen D, Mouton JW. Resistance mechanisms and drug susceptibility testing of nontuberculous mycobacteria. *Drug Resist Updat* 2012;15:149–161.

37 Worlitzsch D, Tarran R, Ulrich M, Schwab U, Cekici A, Meyer KC, Birrer P, Bellon G, Berger J, Weiss T, *et al*. Effects of reduced mucus oxygen concentration in airway *Pseudomonas* infections of cystic fibrosis patients. *J Clin Invest* 2002;109:317–325.

38 Bittar F, Richet H, Dubus JC, Reynaud-Gaubert M, Stremler N, Sarles J, Raoult D, Rolain JM. Molecular detection of multiple emerging pathogens in sputa from cystic fibrosis patients. *PLoS One* 2008; 3:e2908.

39 Brook I, Fink R. Transtracheal aspiration in pulmonary infection in children with cystic fibrosis. *Eur J Respir Dis* 1983;64:51–57.

40 Jewes LA, Spencer RC. The incidence of anaerobes in the sputum of patients with cystic fibrosis. *J Med Microbiol* 1990;31:271–274.

41 Thomassen MJ, Klinger JD, Badger SJ, van Heeckeren DW, Stern RC. Cultures of thoracotomy specimens confirm usefulness of sputum cultures in cystic fibrosis. *J Pediatr* 1984;104:352–356.

42 Tunney MM, Field TR, Moriarty TF, Patrick S, Doering G, Muhlebach MS, Wolfgang MC, Boucher R, Gilpin DF, McDowell A, *et al*. Detection of anaerobic bacteria in high numbers in sputum from patients with cystic fibrosis. *Am J Respir Crit Care Med* 2008;177: 995–1001.

43 Worlitzsch D, Rintelen C, Böhm K, Wollschläger B, Merkel N, Borneff-Lipp M, Döring G. Antibiotic-resistant obligate anaerobes during exacerbations of cystic fibrosis patients. *Clin Microbiol Infect* 2009; 15:454–460.

44 Lambiase A, Catania MR, Rossano F. Anaerobic bacteria infection in cystic fibrosis airway disease. *New Microbiol* 2010;33:185–194.

45 Rogers GB, Carroll MP, Serisier DJ, Hockey PM, Jones G, Kehagia V, Connett GJ, Bruce KD. Use of 16S rRNA gene profiling by terminal restriction fragment length polymorphism analysis to compare bacterial communities in sputum and mouthwash samples from patients with cystic fibrosis. *J Clin Microbiol* 2006;44:2601–2604.

46 Gangell C, Gard S, Douglas T, Park J, de Klerk N, Keil T, Brennan S, Ranganathan S, Robins-Browne R, Sly PD; AREST CF. Inflammatory responses to individual microorganisms in the lungs of children with cystic fibrosis. *Clin Infect Dis* 2011;53:425–432.

47 Tai A, Ranganath S. Anaerobic bacterial infection in patients with cystic fibrosis. *Am J Respir Crit Care Med* 2008;178:994.

48 Sibley CD, Parkins MD, Rabin HR, Duan K, Norgaard JC, Surette MG. A polymicrobial perspective of pulmonary infections exposes an enigmatic pathogen in cystic fibrosis patients. *Proc Natl Acad Sci USA* 2008;105:15070–15075.

49 Ulrich M, Beer I, Braitmaier P, Dierkes M, Kummer F, Krismer B, Schumacher U, Gräpler-Mainka U, Riethmüller J, Jensen PO, *et al*. Relative contribution of *Prevotella intermedia* and *Pseudomonas aeruginosa* to lung pathology in airways of patients with cystic fibrosis. *Thorax* 2010;65:978–984.

50 Tunney MM, Klem ER, Fodor AA, Gilpin DF, Moriarty TF, McGrath SJ, Muhlebach MS, Boucher RC, Cardwell C, Doering G, *et al*. Use of culture and molecular analysis to determine the effect of antibiotic treatment on microbial community diversity and abundance during exacerbation in patients with cystic fibrosis. *Thorax* 2011;66: 579–584.

51 Delhaes L, Monchy S, Fréalle E, Hubans C, Salleron J, Leroy S, Prevotat A, Wallet F, Wallaert B, Dei-Cas E, *et al*. The airway microbiota in cystic fibrosis: a complex fungal and bacterial community—implications for therapeutic management. *PLoS One* 2012;7:e36313.

52 Haase G, Skopnik H, Kusenbach G. *Exophiala dermatitidis* infection in cystic fibrosis. *Lancet* 1990;336:188–189.

53 Haase G, Skopnik H, Groten T, Kusenbach G, Posselt HG. Long-term fungal cultures from sputum of patients with cystic fibrosis. *Mycoses* 1991;34:373–376.

54 Horré R, Schaal KP, Siekmeier R, Sterzik B, de Hoog GS, Schnitzler N. Isolation of fungi, especially *Exophiala dermatitidis*, in patients suffering from cystic fibrosis: a prospective study. *Respiration* 2004; 71:360–366.

55 Kusenbach G, Skopnik H, Haase G, Friedrichs F, Döhmen H. *Exophiala dermatitidis* pneumonia in cystic fibrosis. *Eur J Pediatr* 1992;151: 344–346.

56 Blaschke-Hellmessen R, Lauterbach I, Paul KD, Tintelnot K, Weissbach G. [Detection of *Exophiala dermatitidis* (Kano) De Hoog 1977 in septicemia of a child with acute lymphatic leukemia and in patients with cystic fibrosis]. *Mycoses* 1994;37:89–96.

57 Kondori N, Gilljam M, Lindblad A, Jönsson B, Moore ER, Wennerås C. High rate of *Exophiala dermatitidis* recovery in the airways of patients with cystic fibrosis is associated with pancreatic insufficiency. *J Clin Microbiol* 2011;49:1004–1009.

58 Giraud S, Favennec L, Bougnoux ME, Bouchara JP. *Rasamsonia argillacea* species complex: taxonomy, pathogenesis and clinical relevance. *Future Microbiol* 2013;8:967–978.

59 Chotirmall SH, Al-Alawi M, Mirkovic B, Lavelle G, Logan PM, Greene CM, McElvaney NG. *Aspergillus*-associated airway disease, inflammation, and the innate immune response. *Biomed Res Int* 2013;2013:723129.

60 Chotirmall SH, McElvaney NG. Fungi in the cystic fibrosis lung: bystanders or pathogens? *Int J Biochem Cell Biol* 2014;52: 161–173.

61 Chotirmall SH, O'Donoghue E, Bennett K, Gunaratnam C, O'Neill SJ, McElvaney NG. Sputum *Candida albicans* presages FEV_1 decline and hospital-treated exacerbations in cystic fibrosis. *Chest* 2010;138: 1186–1195.

62 Chotirmall SH, Greene CM, McElvaney NG. *Candida* species in cystic fibrosis: a road less travelled. *Med Mycol* 2010;48:S114–S124.

63 Chotirmall SH, Branagan P, Gunaratnam C, McElvaney NG. *Aspergillus*/allergic bronchopulmonary aspergillosis in an Irish cystic fibrosis population: a diagnostically challenging entity. *Respir Care* 2008;53:1035–1041.

64 Stevens DA, Moss RB, Kurup VP, Knutsen AP, Greenberger P, Judson MA, Denning DW, Crameri R, Brody AS, Light M, *et al*.; Participants in the Cystic Fibrosis Foundation Consensus Conference. Allergic bronchopulmonary aspergillosis in cystic fibrosis—state of the art: Cystic Fibrosis Foundation Consensus Conference. *Clin Infect Dis* 2003;37:S225–S264.

65 Gernez Y, Dunn CE, Everson C, Mitsunaga E, Gudiputi L, Krasinska K, Davies ZA, Herzenberg LA, Tirouvanziam R, Moss RB. Blood basophils from cystic fibrosis patients with allergic bronchopulmonary aspergillosis are primed and hyper-responsive to stimulation by *Aspergillus* allergens. *J Cyst Fibros* 2012;11:502–510.

66 Cohen-Cymberknoh M, Blau H, Shoseyov D, Mei-Zahav M, Efrati O, Armoni S, Kerem E. Intravenous monthly pulse methylprednisolone treatment for ABPA in patients with cystic fibrosis. *J Cyst Fibros* 2009;8:253–257.

67 Elphick HE, Southern KW. Antifungal therapies for allergic bronchopulmonary aspergillosis in people with cystic fibrosis. *Cochrane Database Syst Rev* [serial on the Internet]. 2012 [accessed September 2014];6:CD002204. Available from: http://summaries.cochrane.org/

68 Burgel PR, Baixench MT, Amsellem M, Audureau E, Chapron J, Kanaan R, Honoré I, Dupouy-Camet J, Dusser D, Klaassen CH, *et al*. High prevalence of azole-resistant *Aspergillus fumigatus* in adults with cystic fibrosis exposed to itraconazole. *Antimicrob Agents Chemother* 2012;56:869–874.

69 Rondeau S, Couderc L, Dominique S, Pramil S, Leguillon C, Masseline B, Favennec L, Marguet C. High frequency of voriconazole-related phototoxicity in cystic fibrosis patients. *Eur Respir J* 2012;39:782–784.

70 Tanou K, Zintzaras E, Kaditis AG. Omalizumab therapy for allergic bronchopulmonary aspergillosis in children with cystic fibrosis: a synthesis of published evidence. *Pediatr Pulmonol* 2014;49:503–507.

71 Fillaux J, Brémont F, Murris M, Cassaing S, Rittié JL, Tétu L, Segonds C, Abbal M, Bieth E, Berry A, *et al*. Assessment of *Aspergillus* sensitization or persistent carriage as a factor in lung function impairment in cystic fibrosis patients. *Scand J Infect Dis* 2012;44: 842–847.

72 Baxter CG, Moore CB, Jones AM, Webb AK, Denning DW. IgE-mediated immune responses and airway detection of *Aspergillus* and *Candida* in adult cystic fibrosis. *Chest* 2013;143:1351–1357.

73 Fillaux J, Brémont F, Murris M, Cassaing S, Tétu L, Segonds C, Pipy B, Magnaval JF. *Aspergillus* sensitization or carriage in cystic fibrosis patients. *Pediatr Infect Dis J* 2014;33:680–686.

74 Baxter CG, Dunn G, Jones AM, Webb K, Gore R, Richardson MD, Denning DW. Novel immunologic classification of aspergillosis in adult cystic fibrosis. *J Allergy Clin Immunol* 2013;132:560–566. e510.

75 Amin R, Dupuis A, Aaron SD, Ratjen F. The effect of chronic infection with *Aspergillus fumigatus* on lung function and hospitalization in patients with cystic fibrosis. *Chest* 2010;137:171–176.

76 McMahon MA, Chotirmall SH, McCullagh B, Branagan P, McElvaney NG, Logan PM. Radiological abnormalities associated with *Aspergillus* colonization in a cystic fibrosis population. *Eur J Radiol* 2012;81:e197–e202.

77 Coughlan CA, Chotirmall SH, Renwick J, Hassan T, Low TB, Bergsson G, Eshwika A, Bennett K, Dunne K, Greene CM, *et al*. The effect of *Aspergillus fumigatus* infection on vitamin D receptor expression in cystic fibrosis. *Am J Respir Crit Care Med* 2012; 186:999–1007.

78 Aaron SD, Vandemheen KL, Freitag A, Pedder L, Cameron W, Lavoie A, Paterson N, Wilcox P, Rabin H, Tullis E, *et al*. Treatment of *Aspergillus fumigatus* in patients with cystic fibrosis: a randomized, placebo-controlled pilot study. *PLoS One* 2012;7:e36077.

79 Hassan T, Chotirmall SH, Low TB, Flynn MG, McElvaney NG, Gunaratnam C. Thrombolysis for indwelling catheter related thrombosis and superior vena cava obstruction in cystic fibrosis: a case series. *Ir J Med Sci* 2010;179:469–470.

80 Cimon B, Carrère J, Vinatier JF, Chazalette JP, Chabasse D, Bouchara JP. Clinical significance of *Scedosporium apiospermum* in patients with cystic fibrosis. *Eur J Clin Microbiol Infect Dis* 2000; 19:53–56.

81 Williamson EC, Speers D, Arthur IH, Harnett G, Ryan G, Inglis TJ. Molecular epidemiology of *Scedosporium apiospermum* infection determined by PCR amplification of ribosomal intergenic spacer sequences in patients with chronic lung disease. *J Clin Microbiol* 2001;39:47–50.

82 Tintelnot K, Just-Nübling G, Horré R, Graf B, Sobottka I, Seibold M, Haas A, Kaben U, De Hoog GS. A review of German *Scedosporium prolificans* cases from 1993 to 2007. *Med Mycol* 2009;47:351–358.

83 Summerbell RC, Krajden S, Kane J. Potted plants in hospitals as reservoirs of pathogenic fungi. *Mycopathologia* 1989;106:13–22.

84 Blyth CC, Middleton PG, Harun A, Sorrell TC, Meyer W, Chen SC. Clinical associations and prevalence of *Scedosporium* spp. in Australian cystic fibrosis patients: identification of novel risk factors? *Med Mycol* 2010;48:S37–S44.

85 Defontaine A, Zouhair R, Cimon B, Carrère J, Bailly E, Symoens F, Diouri M, Hallet JN, Bouchara JP. Genotyping study of *Scedosporium apiospermum* isolates from patients with cystic fibrosis. *J Clin Microbiol* 2002;40:2108–2114.

86 Guarro J, Kantarcioglu AS, Horré R, Rodriguez-Tudela JL, Cuenca Estrella M, Berenguer J, de Hoog GS. *Scedosporium apiospermum*: changing clinical spectrum of a therapy-refractory opportunist. *Med Mycol* 2006;44:295–327.