Recent advances in cross-infection in cystic fibrosis: *Burkholderia cepacia* complex, *Pseudomonas aeruginosa*, MRSA and *Pandoraea* spp

Andrew M Jones MRCP A Kevin Webb FRCP

J R Soc Med 2003;**96**(Suppl. 43):66–72 SECTION OF PAEDIATRICS & CHILD HEALTH, 26 NOVEMBER 2002

INTRODUCTION

Cross-infection causes the most concern and discussion amongst cystic fibrosis (CF) health professionals, patients and carers. It causes concern because microbiological status can influence the quality of life and survival of a CF patient.[1–3] The list of bacterial pathogens documented as responsible for cross-infection outbreaks is lengthening and currently includes *Burkholderia cepacia* complex, methicillin-resistant *Staphylococcus aureus* (MRSA), *Pseudomonas aeruginosa* and *Pandoraea* spp.[4–7]

We consider current cross-infection problems in CF and discuss the clinical problems associated with cross-infection, highlighting the major pathogens involved and the consequences of cross-infection for individual patients, specialist centres and the CF community.

CROSS-INFECTION CONTROL AND CF

Cross-infection control in CF requires implementation of basic hygiene measures and cross-infection control principles, taking into account the nature of CF pathogens. Close liaison is needed between the CF multidisciplinary team, microbiologists and infection control teams. Together, they should create a local infection control policy for each CF centre. Good standards of hygiene should be encouraged. Consideration should also be given to minimizing the risk of cross-infection from contamination of the hospital environment and respiratory function equipment. Education for patients, their families and CF healthcare workers is important as their support is essential to the success of cross-infection control policies.

National consensus guidelines for control of MRSA infection in CF have been published.[7] The CF Trust has also produced infection control guidelines for *P. aeruginosa* and *B. cepacia* which can be accessed via the CF Trust website.[8,9]

Manchester Adult Cystic Fibrosis Centre, South Manchester University Hospitals NHS Trust, Southmoor Road, Manchester M23 9LT, UK

Correspondence to: Dr A M Jones

E-mail: andmarkj@hotmail.com

BACTERIAL PATHOGENS ASSOCIATED WITH CROSS-INFECTION IN CF

Bacteria are the most important micro-organisms responsible for the progression of CF lung disease and the prognosis of the patients.[10] The principal bacterial pathogens responsible for documented cross-infection outbreaks at CF centres are described below.

Methicillin-resistant *Staphylococcus aureus* (MRSA)

Staphylococcus aureus is the predominant respiratory pathogen in childhood for those with CF. The upper respiratory tract and nasal passages of healthy humans are commonly colonized by *S. aureus* and most CF pulmonary infections result from endogenous infection with the patient's own organism.[11] Chronic infection of the lower respiratory tract by *S. aureus* can lead to severe lung damage and many CF physicians advocate a policy of long-term prophylactic anti-staphylococcal antibiotics for their patients. This has been shown to be associated with an improved clinical prognosis in neonates,[12] although the effectiveness of this policy beyond childhood years remains controversial.[13]

The majority of strains of *S. aureus* produce penicillinase and therefore penicillinase-stable β-lactam antibiotics, such as methicillin and flucloxacillin, have been the mainstay of treatment of *S. aureus* infections for many years. Strains resistant to these agents, known as methicillin-resistant *S. aureus* (MRSA), emerged in the UK in the 1960s and have increased in incidence over the past few decades.[14,15] Methicillin resistance is through a penicillin binding protein encoded by the *mecA* gene.[14,16]

MRSA strains are now a leading cause of nosocomial infections and several studies suggest that MRSA may be emerging as a community pathogen.[14] MRSA pose greater therapeutic difficulties because of their associated antibiotic resistance. Some strains appear better able to colonize and spread than others; two epidemic lineages (EMRSA) are largely responsible for spread of MRSA in the UK, EMRSA-15 and EMRSA-16.[17] Hospital patients who harbour MRSA can contaminate their environment and inpatient isolation is therefore recommended.[18] Transmission of infection appears to be primarily by person-to-person contact via

contaminated hands, equipment or clothing, although shedding of MRSA (e.g. patients with infected eczema) and airborne spread is also possible.[15]

The prevalence of MRSA among those with CF is increasing and has added to infection control problems faced by CF centres.[19,20] The majority of MRSA infections at the CF centres are thought to be through nosocomial transmission from other hospital patients, rather than transmission between CF patients.[7] Outside of CF, studies have demonstrated that spread of MRSA can be controlled by active surveillance and contact isolation measures.[21] Topical antiseptics and antibiotics can be used for nasal and skin carriage.[7] Active treatment of carriers will often depend on the clinical setting, although this approach has helped to control spread during some MRSA outbreaks.[7]

The clinical significance of MRSA infection in CF is still unclear. A recent study has reported a greater use of intravenous antibiotics and an adverse affect on nutrition and growth for CF children with MRSA infection.[19] Consensus reports recommend adherence to non-CF infection control guidelines for MRSA to help limit spread within CF centres.[7] Attempted eradication of MRSA pulmonary infection using nebulized vancomycin has been advocated for individuals with CF.[22] However, there is no uniform view as to the optimal treatment regime for MRSA in CF patients. Further prospective studies are required to determine the clinical effects of pulmonary MRSA infection and optimal regimen for eradication of MRSA in individuals with CF.

Burkholderia cepacia complex

Chronic *B. cepacia* infection is associated with an increased morbidity and shortened life expectancy for those with CF. The emergence and spread of transmissible strains resulted in a dramatic increase in the incidence and prevalence of *B. cepacia* infection at many CF centres during the past two decades. Patient-to-patient spread occurred both within CF centres[23] and outside during social contact.[5] The subsequent introduction of strict segregation policies has helped to limit *B. cepacia* cross-infection. The recognition of cross-infection, with one *B. cepacia* strain replacing another, often in association with a dramatic worsening in clinical condition of the patient, highlighted the need to further extend cohort segregation of CF patients with *B. cepacia* infection by strain type.[24,25]

The risk of *B. cepacia* cross-infection is related to a number of factors, including patient behaviour, individual host factors,[26] infection control practices at CF centres and the *B. cepacia* strain type.[27] A DNA marker, known as *Burkholderia cepacia* epidemic strain marker (BCESM), has been identified in a number of transmissible strains.[27] The most prevalent transmissible strain in Canadian and UK CF centres, known as the ET12 strain, also possesses a gene (*CbiA*) that encodes for the major structural subunit of unique mucin binding cable pili, which are thought to possibly increase its potential for transmission between CF patients.[27] However, BCESM and *CbiA* are not found in all transmissible strains of the *B. cepacia* complex[28] and genomic fingerprinting of individual isolates is still required for infection control surveillance.

Although the stringent infection control policies have helped to limit cross-infection, they have not entirely eliminated new acquisition of *B. cepacia* complex infection at CF centres. At the Manchester Adult CF Centre there is still a low incidence rate of infection with sporadic strains of *Burkholderia* spp (Figure 1). These are presumed acquired from natural environmental sources, although the source of infectious strains is not known. The recent finding of an epidemic *B. cepacia* complex strain within soil samples[29] has highlighted that human strains are not necessarily distinct from environmental strains. This has added to the ongoing debate over the potential commercial use of *B. cepacia* complex species in agriculture and bioremediation.[2,30] Ongoing work is required to establish if environmental reservoirs constitute a risk for CF patients. Continued microbiological surveillance of *B. cepacia* complex infection is essential to detect future emergence of potentially transmissible strains.

The taxonomy of *B. cepacia* has undergone major revisions during the last few years. Studies have shown that organisms previously identified as *B. cepacia* actually comprise a number of distinct but closely related bacterial species, each known as a genomovar of the *B. cepacia* complex.[31,32] The majority of CF clinical isolates of the *B. cepacia* complex belong to genomovars II (also known as *B. multivorans*) and III.[24,28] The clinical significance of the

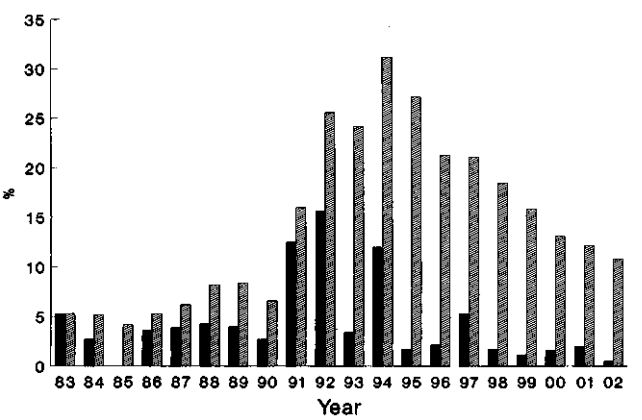

Figure 1 **Incidence and prevalence of infection with *Burkholderia* spp at the Manchester Adult Cystic Fibrosis Centre.** A policy of strict cohort segregation in the mid-1990s has lead to a fall in cases of cross-infection with transmissible *Burkholderia cepacia* genomovar III strains. A low incidence rate persists, mainly as a result of infection by sporadic strains of *Burkholderia* spp from the environment. Striped bars=prevalence; bold bars=incidence

new taxonomic status is still unknown. Whilst most transmissible strains belong to genomovar III *B. cepacia* complex, CF cross-infection outbreaks relating to *B. multivorans*[24,33] and other genomovars[34] have been reported. Ongoing studies are needed to address the relevance of the new taxonomic status to CF clinical care including cross-infection control. The relatively small numbers of CF patients with *B. cepacia* complex infection and potentially large confounding factors between individual patients pose difficulties for such studies.

Some CF patients may transiently acquire infection with a known transmissible strain of *B. cepacia*. When a patient can be declared free of infection, and therefore potentially allowed to cohort with other *B. cepacia*-negative CF patients, can be a difficult clinical dilemma. The CF Trust currently recommends a period of at least 1 year between negative cultures, during which at least three different sputum cultures have been taken, before a patient can be declared as having eradicated a *Burkholderia* sp.[8]

As CF patients travel on holiday in more exotic destinations, they may visit areas where *Burkholderia* spp, including known human pathogens such as *B. mallei* and *B. pseudomallei*, are endemic. Will CF clinicians begin to see cases of infections due to yet other *Burkholderia* spp? Cases of *B. pseudomallei* infection in CF patients visiting South-East Asia have already been described.[35,36] Only continued microbiological surveillance will ensure success of cross-infection control of *Burkholderia* spp in CF.

Pseudomonas aeruginosa

Past and present: evidence for cross-infection

In the 1980s an increase in the incidence and prevalence of multi-resistant *P. aeruginosa* was noted in the Danish CF Centre.[37] However, the phenotypic bacterial typing systems available at the time were unable to provide compelling evidence that a single strain was responsible. As the majority of other early studies did not find evidence of significant *P. aeruginosa* cross-infection among patients at CF centres[38–40] or holiday camps[41,42] most clinicians and microbiologists concluded that *P. aeruginosa* cross-infection was not a significant problem in CF.

Recently opinions have changed. Studies using molecular fingerprinting of *P. aeruginosa* isolates have demonstrated convincing evidence of clonal spread at CF holiday camps in Europe[43,44] and specialist centres in the UK and Australia.[4,45–47] Another recent study at the Vancouver CF Centre using randomly amplified polymorphic DNA typing did not reveal any evidence of significant cross-infection with *P. aeruginosa*.[48] The conclusion is that *P. aeruginosa* cross-infection does exist at some but not all CF centres. Only widespread microbiological surveillance will reveal the true extent of this problem at all UK CF centres. Whilst isolates of transmissible *P. aeruginosa* strains may exhibit unusual phenotypic features, including antibiotic resistance, none of these features can be regarded as discriminatory and microbiological surveillance for *P. aeruginosa* cross-infection should involve some form of bacterial genotyping to identify individual strains.

Why has *P. aeruginosa* cross-infection emerged?

It is known that the particular strain of *P. aeruginosa* may be important in the establishment of chronic pulmonary infection in people with CF. Elegant microbiological studies have demonstrated that the degree of chemotaxis of *P. aeruginosa* to the major sugar and amino acid components of mucin is strain specific.[49] Findings from an outbreak of *P. aeruginosa* infection related to a hydrotherapy pool highlighted an increased predilection of a particular *P. aeruginosa* clone, which possessed the highest mucinophilic and chemotactic properties of all the different *P. aeruginosa* strains isolated from the pool water, for the CF lung.[3]

The versatility and adaptability of *P. aeruginosa* is reflected in the immense size and sophistication of its genome which contains 6.3 million base pairs.[50] Isolates of *P. aeruginosa* from CF hosts show a remarkably high rate of mutation which, it has been suggested, allows the continued and rapid adaptation of the bacterial population and helps to ensure their continued survival within CF lungs.[51] The recently reported cross-infection outbreaks seem to be as result of the spread of particular transmissible strains between patients, rather than a simple breakdown in infection control measures at the CF centres. Many isolates of transmissible *P. aeruginosa* strains exhibit unusual phenotypic features,[4,47] super-infection has also been reported.[45] The reasons behind the emergence of these newly identified strains and factors associated with their transmissibility, however, are at present unclear.

What is the mechanism of cross-infection?

The mechanism of *P. aeruginosa* cross-infection is not known and establishing experimental proof of the mode(s) of spread is prevented by ethical limitations. Recent environmental screening at the Manchester Adult CF Centre during a documented cross-infection outbreak has demonstrated airborne dissemination of transmissible *P. aeruginosa* by CF patients.[52] No other reservoir of infection was found in the inanimate environment. These findings suggest that the mode of cross-infection may be patient-to-patient spread of *P. aeruginosa* by airborne dissemination. This raises implications for infection control policies implying measures may need to include cohort isolation of patients who harbour transmissible *P. aeruginosa*. This will further increase pressure on the limited resources

of many large CF centres (Box 1). Whichever measures are implemented, continued microbiological surveillance must be undertaken to determine their outcome.

Clinical effects of *P. aeruginosa* cross-infection

> 'The success of early identification and treatment in preventing Pseudomonas infection becoming established and chronic determines the patient's future quality of life and long-term survival'—Cystic Fibrosis Trust, 2003.[53]

The acquisition of *P. aeruginosa* infection is associated with an increase in morbidity and mortality for individuals with CF. Once chronic infection becomes established, it is virtually impossible to eradicate. The prevention of chronic *P. aeruginosa* infection in CF relies on cross-infection control and the eradication of early infection. The emergence of transmissible strains of *P. aeruginosa* at CF centres is a concern as they expose other *P. aeruginosa*-negative patients to an increased risk of acquisition of infection. Eradication regimens for early *P. aeruginosa* infection remain an important tool in the armoury of the CF team in the management of their patients.[9,53,54] The potential ability of transmissible multi-resistant *P. aeruginosa* to resist eradication[4] and progress to chronic infection is of further concern.

What are the effects of cross-infection for CF patients who already harbour sporadic strains of *P. aeruginosa*? Two recent studies have reported an increase in morbidity for CF patients who harbour transmissible *P. aeruginosa* above that of those with infection by sporadic strains.[46,55] Patients infected with transmissible strains were found to have a greater number of respiratory exacerbations, a greater intensity of intravenous antibiotic treatment and inpatient treatment. Although there are resource implications in the implementation of infection control measures, there may be potential cost implications for ignoring spread of transmissible multi-resistant *P. aeruginosa*.

The transmissibility of a CF pathogen however, should not be confused with its virulence. At the Manchester Adult CF Centre, a recent cross-sectional study failed to show any differences between levels of inflammatory markers in clinically stable adult CF patients with infection by transmissible and sporadic *P. aeruginosa*. The virulence of transmissible *P. aeruginosa* is currently unknown but warrants further study. Furthermore, the importance of balancing the considerable benefits of centre care against the small risk of *P. aeruginosa* infection cannot be understated: centre care is associated with a better outcome for CF patients,[56] even during a cross-infection outbreak.[57]

Box 1 **Current outpatient clinics at the Manchester Adult Cystic Fibrosis Centre: cohort segregation by organism**

Staphylococcus aureus, *Haemophilus influenzae*
Methicillin-resistant *S. aureus* (MSRA)
Pseudomonas aeruginosa—sporadic strains
Pseudomonas aeruginosa—transmissible strains
Burkholderia gladioli
Burkholderia multivorans
Other non-genomovar III *Burkholderia cepacia* complex
Burkholderia cepacia complex genomovar III—sporadic strains
Burkholderia cepacia complex genomovar III—transmissible strains

Other organisms

Other bacteria such as *Stenotrophomonas maltophilia*, *Alcaligines xylosoxidans*, *Ralstonia pickettii* and *Proteus* spp may occasionally cause chronic pulmonary colonization in CF patients, although their pathogenic role in CF pulmonary disease is unclear. Some develop pulmonary infections with *atypical mycobacteria*. At present, cross-infection with these bacteria has not been reported at CF centres. Furthermore, the rate of transmission of *S. maltophilia* between CF siblings seems to be low.[58]

Pandoraea species

Recently, using isolates identified from the environment and CF sputa, taxonomists have characterized a novel genus, *Pandoraea* spp, containing five species.[6] These organisms appear to be potential pathogens for individuals with CF. Cross-infection with *Pandoraea* spp has already been reported at CF Centres outside the UK.[6] Consideration should be given to microbiological surveillance to investigate the potential spread of *Pandoraea* spp to other CF centres.

RESPIRATORY VIRUSES

Respiratory viruses, such as adenoviruses, respiratory syncytial virus and influenza viruses A and B are responsible for some of the acute exacerbations of the pulmonary disease in CF and can cause a dramatic fall in pulmonary function.[59] Attention should therefore be paid to attempts to minimize the risk of spread of respiratory viruses to CF patients. Similarly, individuals with CF are at risk of severe varicella pneumonitis and exposure carrying a risk of infection should be avoided; in cases of potential exposure, consideration should be given to early prophylactic treatment with aciclovir.[60]

SOCIAL AND PSYCHOLOGICAL CONSEQUENCES OF CROSS-INFECTION

There can be few more emotive articles ever published in the medical press than the moving story of two brothers with CF who decided to live apart when one developed *B. cepacia* infection.[61]

The implementation of strict segregation policies to limit the spread of transmissible pathogens has been at a price to the previous closely-knit structure of the CF community. Holiday camps are now discouraged. Individuals who harbour transmissible or antibiotic resistant organisms are advised not to attend CF meetings and social gatherings. Patient comments from the annual user satisfaction survey at the Manchester Adult Centre give examples of the psychosocial implications of cross-infection in CF (Box 2). Some patients raise concerns about the potential of cross-infection, whilst patients with *B. cepacia* infection complain of the restrictions imposed by segregation policies and express feelings of stigmatization and isolation. Paediatric CF patients list cross-infection as a major concern at the time of transfer of their care to adult centres.[62] Effective communication between the CF team and patients and their relatives is necessary to attempt to ease anxiety among all patients. The team should encourage maintenance of cross-infection control policies, whilst providing psychosocial support to individual patients who may feel stigmatized.[63] Ways of allowing patients to communicate and interact whilst minimizing the risk of cross-infection, such as use of the Internet and the provision of teleconferencing facilities, should also be explored.

MICROBIOLOGICAL SURVEILLANCE FOR CROSS-INFECTION

Microbiological surveillance for cross-infection with the major CF bacterial pathogens should include genomic fingerprinting of isolates. The method chosen must have suitable discriminatory power for the species under investigation. The different molecular bacterial fingerprinting techniques available are beyond the scope of this clinical review. Antibiograms are unreliable for assessing clonality of isolates and cannot be used for surveillance of cross-infection. Genomic fingerprinting techniques can be time consuming. The potentially high number of isolates needed to be screened, such as *P. aeruginosa* at large adult centres, can present additional difficulties for microbiological surveillance.

Specialist CF microbiological reference laboratories have been developed to assist clinicians and microbiological laboratories in the identification, epidemiology and surveillance of the major CF pathogens. The CF Trust helps fund two UK reference laboratories, based in London and Edinburgh.[9]

Recently, the genomes for *P. aeruginosa*, *B. cepacia* genomovar III and MRSA have been sequenced.[50,64] The rapid advances being made in microarray and DNA chip technologies may allow investigators to use comparative genomics to identify genes associated with transmissibility. Whether this will translate into the development of molecular bedside tests to help clinicians identify an organism and indicate its likely potential for spread to help guide infection control remains to be seen.

Box 2 **Psychosocial issues in cystic fibrosis (CF) cross-infection: patient comments from a recent user satisfaction survey at the Manchester Adult CF Centre.** Comments raised include concern of potential cross-infection voiced by some non-*Burkholderia cepacia* infected CF patients; patients with *B. cepacia* infection comment on the difficult restrictions due to segregation and feelings of stigmatization and isolation

'Being treated different to non-*cepacia* patients'
'Some concern about cross-infection'
'There isn't a choice for people with *cepacia*'
'Bit worried about cross-infection'
'Have more choice of times for *cepacia* patients, as it is difficult to keep coming on the same day and time, when needing to work'
'Concerned with cross-infection'
'Segregated patients need informing of what is going on in other wards as they can feel isolated, in the dark and also in two-star accommodation'
'I feel that it is not good to have *Pseudomonas* (clinics) on the same day as *cepacia* as there may be a slight chance of the germ in the air'

CONCLUSION

Cross-infection is important in CF as the microbiological status of a patient influences their quality of life, opportunities for social contact with peers, future prospects for transplantation, disease progression and life expectancy. The introduction of strict segregation policies has helped to limit spread of transmissible strains of the *B. cepacia* complex. The emerging prevalence of transmissible strains of other CF pathogens such as MRSA and *P. aeruginosa* presents further challenges for cross-infection control. Success in limiting spread will require adequate provision of resources, continued microbiological surveillance and close liaison between infection control teams, microbiologists and CF multi-disciplinary teams.

Acknowledgments We should like to thank Mary Dodd and Rowland Bright-Thomas, Manchester Adult CF Centre, for their helpful comments and Professor John Govan and Dr Cathy Doherty, Medical Microbiology, University of Edinburgh, for their microbiological expertise.

REFERENCES

1 CF Foundation. *Patient Registry 1999: Annual Report*, Bethesda: CF Foundation, 2000

2 Jones AM, Dodd ME, Webb AK. *Burkholderia cepacia*: current clinical issues, environmental controversies and ethical dilemmas. *Eur Respir J* 2001;**17**:295–301

3 Govan JR, Deretic V. Microbial pathogenesis in cystic fibrosis: mucoid *Pseudomonas aeruginosa* and *Burkholderia cepacia*. *Microbiol Rev* 1996;**60**:539–74

4 Jones AM, Govan JR, Doherty CJ, *et al*. Spread of a multiresistant strain of *Pseudomonas aeruginosa* in an adult cystic fibrosis clinic. *Lancet* 2001;**358**:557–8

5 Govan JR, Brown PH, Maddison J, *et al*. Evidence for transmission of *Pseudomonas cepacia* by social contact in cystic fibrosis. *Lancet* 1993;**342**:15–19

6 Coenye T, Liu L, Vandamme P, LiPuma JJ. Identification of pandoraea species by 16S ribosomal DNA-based PCR assays. *J Clin Microbiol* 2001;**39**:4452–5

7 Report M. MRSA in cystic fibrosis. London, 16 June 1997. *J Hosp Infect* 1998;**40**:179–91

8 Group CFTIC. *A statement on* Burkholderia cepacia. London: Cystic Fibrosis Trust, 1999

9 Group CFTCoI. Pseudomonas aeruginosa *infection in people with cystic fibrosis*. London: Cystic Fibrosis Trust, 2001

10 Govan JR, Nelson JW. Microbiology of lung infection in cystic fibrosis. *Br Med Bull* 1992;**48**:912–30

11 Govan JR. Infection control in cystic fibrosis: methicillin-resistant *Staphylococcus aureus*, *Pseudomonas aeruginosa* and the *Burkholderia cepacia* complex. *J R Soc Med* 2000;**93**:40–5

12 Weaver LT, Green MR, Nicholson K, *et al*. Prognosis in cystic fibrosis treated with continuous flucloxacillin from the neonatal period. *Arch Dis Child* 1994;**70**:84–9

13 Smyth A, Walters S. Prophylactic antibiotics for cystic fibrosis. *Cochrane Database Syst Rev* 2000;**2**:CD001912

14 Oliveira DC, Tomasz A, de Lencastre H. Secrets of success of a human pathogen: molecular evolution of pandemic clones of meticillin-resistant *Staphylococcus aureus*. *Lancet Infect Dis* 2002;**2**:180–9

15 Report WP. Revised guidelines for the control of methicillin-resistant *Staphylococcus aureus* infection in hospitals. *J Hosp Infect* 1998;**39**:253–90

16 Kreiswirth B, Kornblum J, Arbeit RD, *et al*. Evidence for a clonal origin of methicillin resistance in *Staphylococcus aureus*. *Science* 1993;**259**:227–30

17 Enright MC, Day NP, Davies CE, Peacock SJ, Spratt BG. Multilocus sequence typing for characterization of methicillin-resistant and methicillin-susceptible clones of *Staphylococcus aureus*. *J Clin Microbiol* 2000;**38**:1008–15

18 Boyce JM, Potter-Bynoe G, Chenevert C, King T. Environmental contamination due to methicillin-resistant *Staphylococcus aureus*: possible infection control implications. *Infect Control Hosp Epidemiol* 1997;**18**:622–7

19 Miall LS, McGinley NT, Brownlee KG, Conway SP. Methicillin resistant *Staphylococcus aureus* (MRSA) infection in cystic fibrosis. *Arch Dis Child* 2001;**84**:160–2

20 Thomas SR, Gyi KM, Gaya H, Hodson ME. Methicillin-resistant *Staphylococcus aureus*: impact at a national cystic fibrosis centre. *J Hosp Infect* 1998;**40**:203–9

21 Farr BM, Salgado CD, Karchmer TB, Sheretz RJ. Can antibiotic-resistant nosocomial infections be controlled? *Lancet Infect Dis* 2001;**i**:38–45

22 Maiz L, Canton R, Mir N, Baquero F, Escobar H. Aerosolized vancomycin for the treatment of methicillin-resistant *Staphylococcus aureus* infection in cystic fibrosis. *Pediatr Pulmonol* 1998;**26**:287–9

23 LiPuma JJ, Dasen SE, Nielson DW, Stern RC, Stull TL. Person-to-person transmission of *Pseudomonas cepacia* between patients with cystic fibrosis. *Lancet* 1990;**336**:1094–6

24 Mahenthiralingam E, Vandamme P, Campbell ME, *et al*. Infection with *Burkholderia cepacia* complex genomovars in patients with cystic fibrosis: virulent transmissible strains of genomovar III can replace *Burkholderia multivorans*. *Clin Infect Dis* 2001;**33**:1469–75

25 Ledson MJ, Gallagher MJ, Corkill JE, Hart CA, Walshaw MJ. Cross infection between cystic fibrosis patients colonised with *Burkholderia cepacia*. *Thorax* 1998;**53**:432–6

26 Garred P, Pressler T, Madsen HO, *et al*. Association of mannose-binding lectin gene heterogeneity with severity of lung disease and survival in cystic fibrosis. *J Clin Invest* 1999;**104**:431–7

27 Mahenthiralingam E, Simpson DA, Speert DP. Identification and characterization of a novel DNA marker associated with epidemic *Burkholderia cepacia* strains recovered from patients with cystic fibrosis. *J Clin Microbiol* 1997;**35**:808–16

28 LiPuma JJ, Spilker T, Gill LH, Campbell PW, 3rd, Liu L, Mahenthiralingam E. Disproportionate distribution of *Burkholderia cepacia* complex species and transmissibility markers in cystic fibrosis. *Am J Respir Crit Care Med* 2001;**164**:92–6

29 LiPuma JJ, Spilker T, Coenye T, Gonzalez CF. An epidemic *Burkholderia cepacia* complex strain identified in soil. *Lancet* 2002;**359**:2002–3

30 Holmes A, Govan J, Goldstein R. Agricultural use of *Burkholderia (Pseudomonas) cepacia*: a threat to human health? *Emerg Infect Dis* 1998;**4**:221–7

31 Mahenthiralingam E, Coenye T, Chung JW, *et al*. Diagnostically and experimentally useful panel of strains from the *Burkholderia cepacia* complex. *J Clin Microbiol* 2000;**38**:910–13

32 Vandamme P, Holmes B, Vancanneyt M, *et al*. Occurrence of multiple genomovars of *Burkholderia cepacia* in cystic fibrosis patients and proposal of *Burkholderia multivorans* sp nov. *Int J Syst Bacteriol* 1997;**47**:1188–200

33 Segonds C, Heulin T, Marty N, Chabanon G. Differentiation of burkholderia species by PCR-restriction fragment length polymorphism analysis of the 16S rRNA gene and application to cystic fibrosis isolates. *J Clin Microbiol* 1999;**37**:2201–8

34 Agodi A, Mahenthiralingam E, Barchitta M, Giannino V, Sciacca A, Stefani S. *Burkholderia cepacia* complex infection in Italian patients with cystic fibrosis: prevalence, epidemiology, and genomovar status. *J Clin Microbiol* 2001;**39**:2891–6

35 Visca P, Cazzola G, Petrucca A, Braggion C. Travel-associated *Burkholderia pseudomallei* infection (Melioidosis) in a patient with cystic fibrosis: a case report. *Clin Infect Dis* 2001;**32**:E15–16

36 Schulin T, Steinmetz I. Chronic melioidosis in a patient with cystic fibrosis. *J Clin Microbiol* 2001;**39**:1676–7

37 Pedersen SS, Koch C, Hoiby N, Rosendal K. An epidemic spread of multiresistant *Pseudomonas aeruginosa* in a cystic fibrosis centre. *J Antimicrob Chemother* 1986;**17**:505–16

38 Grothues D, Koopmann U, von der Hardt H, Tummler B. Genome fingerprinting of *Pseudomonas aeruginosa* indicates colonization of cystic fibrosis siblings with closely related strains. *J Clin Microbiol* 1988;**26**:1973–7

39 Kelly NM, Fitzgerald MX, Tempany E, O'Boyle C, Falkiner FR, Keane CT. Does pseudomonas cross-infection occur between cystic-fibrosis patients? *Lancet* 1982;**ii**:688–90

40 Speert DP, Campbell ME. Hospital epidemiology of *Pseudomonas aeruginosa* from patients with cystic fibrosis. *J Hosp Infect* 1987;**9**:11–21

41 Speert DP, Lawton D, Damm S. Communicability of *Pseudomonas aeruginosa* in a cystic fibrosis summer camp. *J Pediatr* 1982;**101**:227–8

42 Smith DL, Smith EG, Gumery LB, Stableforth DE, Dalla Costa LM, Pitt TL. Epidemiology of *Pseudomonas aeruginosa* infection in cystic fibrosis and the use of strain genotyping. *J Infect* 1993;**26**:325–31

43 Ojeniyi B, Frederiksen B, Hoiby N. *Pseudomonas aeruginosa* cross-infection among patients with cystic fibrosis during a winter camp. *Pediatr Pulmonol* 2000;**29**:177–81

44 Hunfeld KP, Schmidt C, Krackhardt B, *et al*. Risk of *Pseudomonas aeruginosa* cross-colonisation in patients with cystic fibrosis within a holiday camp—a molecular–epidemiological study. *Wien Klin Wochenschr* 2000;**112**:329–33

45 McCallum SJ, Corkill J, Gallagher M, Ledson MJ, Hart CA, Walshaw MJ. Superinfection with a transmissible strain of *Pseudomonas aeruginosa* in adults with cystic fibrosis chronically colonized by *P. aeruginosa*. *Lancet* 2001;**358**:558–60

46 Armstrong DS, Nixon GM, Carzino R, *et al*. Detection of a widespread clone of *Pseudomonas aeruginosa* in a pediatric cystic fibrosis clinic. *Am J Respir Crit Care Med* 2002;**166**:983–7

47 Cheng K, Smyth RL, Govan JR, *et al*. Spread of beta-lactam-resistant *Pseudomonas aeruginosa* in a cystic fibrosis clinic. *Lancet* 1996;**348**:639–42

48 Speert DP, Campbell ME, Henry DA, *et al*. Epidemiology of *Pseudomonas aeruginosa* in cystic fibrosis in British Columbia, Canada. *Am J Respir Crit Care Med* 2002;**166**:988–93

49 Nelson JW, Tredgett MW, Sheehan JK, Thornton DJ, Notman D, Govan JR. Mucinophilic and chemotactic properties of *Pseudomonas aeruginosa* in relation to pulmonary colonization in cystic fibrosis. *Infect Immun* 1990;**58**:1489–95

50 Stover CK, Pham XQ, Erwin AL, *et al*. Complete genome sequence of *Pseudomonas aeruginosa* PA01, an opportunistic pathogen. *Nature* 2000;**406**:959–64

51 Oliver A, Canton R, Campo P, Baquero F, Blazquez J. High frequency of hypermutable *Pseudomonas aeruginosa* in cystic fibrosis lung infection. *Science* 2000;**288**:1251–4

52 Jones AM, Dodd M, Doherty C, *et al*. Identification of airborne dissemination of epidemic multi-resistant strains of *Pseudomonas aeruginosa* at a CF centre during a cross-infection outbreak. *Pediatr Pulmonol* 2002;**S24**:295

53 Group CFTA. *Antibiotic Treatment of Cystic Fibrosis*. London: Cystic Fibrosis Trust, 2000

54 Frederiksen B, Koch C, Hoiby N. Antibiotic treatment of initial colonization with *Pseudomonas aeruginosa* postpones chronic infection and prevents deterioration of pulmonary function in cystic fibrosis. *Pediatr Pulmonol* 1997;**23**:330–5

55 Jones AM, Dodd ME, Doherty CJ, Govan JRW, Webb AK. Increased treatment requirements of patients with cystic fibrosis who harbour a highly transmissible strain of *Pseudomonas aeruginosa*. *Thorax* 2002;**57**:924–5

56 Mahadeva R, Webb K, Westerbeek RC, *et al*. Clinical outcome in relation to care in centres specialising in cystic fibrosis: cross sectional study. *BMJ* 1998;**316**:1771–5

57 Zimakoff J, Hoiby N, Rosendal K, Guilbert JP. Epidemiology of *Pseudomonas aeruginosa* infection and the role of contamination of the environment in a cystic fibrosis clinic. *J Hosp Infect* 1983;**4**:31–40

58 Demko CA, Stern RC, Doershuk CF. *Stenotrophomonas maltophilia* in cystic fibrosis: incidence and prevalence. *Pediatr Pulmonol* 1998;**25**:304–8

59 Conway SP, Simmonds EJ, Littlewood JM. Acute severe deterioration in cystic fibrosis associated with influenza A virus infection. *Thorax* 1992;**47**:112–14

60 Ong EL, Mulvenna P, Webb KA. Varicella-zoster infection in adults with cystic fibrosis: role of acyclovir. *Scand J Infect Dis* 1991;**23**:283–5

61 Webb AK. Two memorable patients. *BMJ* 2000;**320**:1324

62 Boyle MP, Farukhi Z, Nosky ML. Strategies for improving transition to adult cystic fibrosis care, based on patient and parent views. *Pediatr Pulmonol* 2001;**32**:428–36

63 Duff AJ. Psychological consequences of segregation resulting from chronic *Burkholderia cepacia* infection in adults with CF. *Thorax* 2002;**57**:756–8

64 Kuroda M, Ohta T, Uchiyama I, *et al*. Whole genome sequencing of meticillin-resistant *Staphylococcus aureus*. *Lancet* 2001;**357**:1225–40