Airborne dissemination of *Burkholderia* (*Pseudomonas*) *cepacia* from adult patients with cystic fibrosis

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Abstract

Background – *Burkholderia* (*Pseudomonas*) *cepacia* is an increasingly important pathogen in patients with cystic fibrosis but it is unclear how it spreads from patient to patient. A study was undertaken to determine whether *B cepacia* could be recovered from room air occupied by colonised adult patients with cystic fibrosis.

Methods – Air samples were obtained consecutively from an enclosed room or isolation cubicle before, during, and after occupation by six patients on nine occasions using a surface air sampler incorporating contact plates with selective medium. Settle plates were also used and sputum from five patients was cultured.

Results – *B cepacia* was recovered from room air during occupation by five of six patients, the number of bacteria ranging from 1 to 158 cfu/m³ (mean 32 cfu/m³). The number of bacteria isolated was greater when patients were coughing. *B cepacia* persisted in room air on four occasions after the patient left the room, on one occasion for up to 45 minutes.

Conclusions – The isolation of *B cepacia* from the air of rooms occupied by colonised patients suggests that dissemination might occur by aerosol as well as by direct physical contact with patients or contaminated environmental sites.

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There is increasing interest in *Burkholderia* (*Pseudomonas*) *cepacia*^{1,2} as a cause of infection in patients with cystic fibrosis in North America and Europe.³ Acquisition by patients with cystic fibrosis may follow three clinical courses: long term colonisation without deterioration in pulmonary function, chronic infection, or acute fulminating disease with rapidly deteriorating lung function and death within weeks.⁴ Risk factors for acquisition include severe pulmonary disease, recent admission to hospital, aerosol therapy, and a *B cepacia* infected sibling.^{3,4} The pathogenicity of this organism in man is poorly understood and many doubt its ability to cause disease. Pre-existing lung damage and the effects of other pathogens may enhance its pathogenic potential,⁵ although in a recent study of 13 patients colonised by *B cepacia* only eight (62%) were also colonised with *Pseudomonas aeruginosa*.⁶

There is considerable anxiety amongst patients, their families, and physicians that person-to-person transmission may occur. Patients may contaminate the immediate environment resulting in indirect transmission.⁷ Ribotyping strongly implicates person-to-person spread,⁸ and the question arises whether positive patients should be segregated from other patients. We report the airborne dissemination of *B cepacia* from colonised patients with cystic fibrosis.

Methods

PATIENTS

Colonisation of patients with cystic fibrosis in Nottingham by *B cepacia* was first confirmed in November 1991 and now involves one third (16/48) of adult patients. Six colonised adult patients were studied (three on two occasions). Patients were chosen on the basis of availability and willingness to participate, but were representative of all colonised patients. The subjects were also persistently colonised with *Ps aeruginosa*. There were four men and two women, and the mean age was 22.7 years (range 20-27). All had progressive lung deterioration before isolation of *B cepacia* with a forced expiratory volume in one second (FEV₁) of 36.3% predicted and a mean forced vital capacity (FVC) of 52.7%. Two of the patients were awaiting lung transplantation while four were on one or more intravenous antibiotics and none was receiving nebulised antibiotics.

MICROBIOLOGY

Sputum culture

Sputum from five of the six patients was inoculated on to *B cepacia* selective medium (MAST, UK) incorporating ticarcillin and polymyxin B to achieve a final concentration of 100 mg/l and 300 000 units/l, respectively. Agar plates were incubated for 40 hours at 37°C. Isolates were confirmed as *B cepacia* in conjunction with the Division of Hospital Infection, Central Public Health Laboratory if they were resistant to polymyxin B, oxidative in Hugh and Leifson's medium, produced cytochrome oxidase, did not produce arginine dihydrolase, and grew on Simmons' citrate medium.⁹

Environmental sampling

A surface air system (SAS) air sampler (Cherwell Laboratories, UK)) positioned 100 cm

Air counts (colony forming units/m³) of *Burkholderia cepacia*

Patient no.	Counts (room occupied)*	Counts (room empty)	Settle plates
1	(1) 1-7 (2) 1-2	(1) Positive at 0, 15 min (2) All negative	(1) Negative (2) Negative
2	2-3	All negative	Negative
3	All negative	All negative	Negative
4	1-3	All negative	Negative
5	(1) 23-97 (2) 27-121	(1) Positive at 0, 15 and 30 min (2) Positive at 0, 15, 30 and 45 min	(1) Positive (2) Not done
6	(1) 105-158 (2) 2-7	(1) Positive at 0 min (2) All negative	(1) Negative (2) Positive

* Range during coughing and whilst at rest.

from the patient was used to sample 900 l of air over a five minute period before and during occupation of an enclosed room (2.2 m × 3 m × 2.5 m or an isolation cubicle (3.3 m × 3 m × 2.7 m) by patients while coughing and while at rest. Air samples were taken at 15 minute intervals for one hour after the patient had vacated the room and also 18 hours later. There was at least one week between sampling of room air occupied by different patients. The SAS sampler aspirates air by a fan at a fixed speed (180 l/min) to a maximum of 900 l over five minutes. Air was directed on to the agar surface of a contact plate containing *B cepacia* selective medium as described above. Contact plates were incubated in air at 30°C for up to seven days and the number of colonies counted and corrected for the coincidence of two or more colony forming units (cfu) passing through the same hole.¹⁰ The number of bacteria isolated was expressed as cfu/m³ of air.

Settle plates, also incorporating *B cepacia* medium, were placed for 18 hours in the immediate vicinity of patients on all but one occasion.

Results

B cepacia was recovered from room air during occupation by five of the six patients (table). All samples taken before occupation were negative. Sputum was unavailable for culture from patient 3 at the time of the study but subsequent samples have been positive. The amount of *B cepacia* recovered from positive room air ranged from 1 to 158 cfu/m³ with a mean of 32 cfu/m³. Twenty three of 25 air samples obtained while patients were coughing and eight of 12 taken when not coughing were positive. The mean count during coughing was 40 cfu/m³ compared with 15 cfu/m³ of air samples at rest.

Following exit of the patient from the room, *B cepacia* was recovered on four occasions (three patients, table). In two patients samples were positive at 15 and/or 30 minutes. Counts ranged from 3 to 53 cfu/m³ of air. Air counts taken from the enclosed room occupied by patient 5 (second occasion) ranged from 27 to 121 cfu/m³ and a sample was also positive 45 minutes after departure. Settle plates were positive on two occasions (patients 5 and 6).

Discussion

B cepacia was present in the air of a closed room or isolation cubicle during and following occupation by patients with cystic fibrosis and persisted for up to 45 minutes afterwards. All

air samples were negative before occupation and it is assumed that airborne bacteria originated from the patients. This suggests that airborne contamination by *B cepacia* is possible and may be a mode of transmission in addition to person-to-person spread by physical contact with colonised patients or contaminated surfaces.

Some of our patients were receiving antibiotics for the treatment of *Ps aeruginosa* infection and the impact of this on dissemination is unclear because *B cepacia* in Nottingham is resistant to gentamicin, tobramycin, azlocillin, aztreonam, imipenem, and ciprofloxacin. Combinations of antimicrobial agents may result in lower counts of *B cepacia* because of synergistic activity. Alternatively, antibiotics may have selected for *B cepacia* and assisted in dissemination by reducing other respiratory flora. Quantitative sputum cultures, which were not carried out in this study, might clarify the effect of antibiotic treatment.

The importance of air in the transmission of infection is well recognised but the sampling of hospital air for microbiological purposes is only indicated as part of operating theatre commissioning during an outbreak of infection or for research. Accurate methods include slit air samplers or the SAS sampler which are convenient and acceptable for most purposes.^{10,11} Settle plates are a crude, if cheap, method and may not correlate with results from an SAS sampler as was the case for patient 6.

A retrospective case-control study has implicated the use of nebuliser or humidifier therapy in nosocomial acquisition as *B cepacia* was isolated from nebuliser reservoirs although air samples were negative.¹² The spread of *B cepacia* amongst patients with cystic fibrosis attending two centres in the UK was documented using bacteriocin and molecular typing.⁶ The organism was isolated from the hands of three patients and disposable spirometry equipment.⁶ The authors of both studies recommend patient segregation for hospital inpatients and outpatients. The failure to isolate *B cepacia* from air samples in these studies may be due to differences between the patients and the circumstances and frequency of monitoring air, and the technique used. Slit samplers as used in these two studies operate at an air flow of 30 l/min whereas the SAS system can sample at 180 l/min.

B cepacia has been isolated from equipment, shower drains, vase water, and plant soil,^{3,4} which probably reflects contamination from heavily colonised patients. The survival of this organism, especially in moist environments, may also be a factor, and transmission in saliva

via direct contact (kissing) or shared vessels or eating utensils is possible. The involvement of many adult cystic fibrosis patients in self-help and social groups means that frequent and regular meetings occur and close, even intimate, contact may arise. Consequently, patient-to-patient transmission via direct contact, contact with contaminated environmental sources, or possibly by the airborne route must be considered here as well as in hospital.

Physical separation in hospital of patients colonised with *B cepacia* in their sputum has led to a decrease in the incidence of this organism in patients with cystic fibrosis.¹³ *Burkholderia cepacia* positive patients should avoid kissing and other intimate contact with non-colonised patients.¹⁴ We have segregated patients since May 1992, but guidelines to minimise the transmission of *B cepacia* must take account of social and emotional factors and avoid making patients feel like outcasts.¹⁵ The findings that *B cepacia* may contaminate room air suggests that segregation whilst in hospital should be continued where possible.

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