## 96 Microbiological audit in Cystic Fibrosis Romanian children

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**Aim of study:** to evaluate the spectrum of pulmonary infection among Romanian CF children.
**Materials and Methods:** study lot included 93 children, median age at diagnosis = 11.8 yrs, ranging from 3 month to 18 yr. Patients were classified in 4 age groups, as follow: under 1 years old (n = 18), 1–3 yrs (n = 34), 3–7 yrs (n = 7), 7–18 years old (n = 33). We have analyzed microbiological results obtained from: sputum samples, throat swabs or pharingotracheal aspirates, for five years ago. The pursued pathogens were: *P. aeruginosa*, *S. aureus*, MRSA, *E. coli*, Klebsiella sp, *S. maltophilia*, *Aspergilus fumigatus*, *Candida albicans*. We could not determine Burkholderia cepacia complex.
**Results:** Table 1-Pathogens prevalence in CF children. Among study lot 19.35% (18 patients) had infection with 2 or more pathogens.
**Conclusions:** *P. aeruginosa* is the most frequent pathogen in our lot study and its prevalence is rising with age, while *S. aureus* predominates in babies. A.fumigatus has is a significant pathogen, also in older children. An important percentage of mixed infection was registered, especially in delayed diagnosed children.

Table 1. Pathogens prevalence

| Pathogens/Age group | 3 mo-1 yr | 1–3 yrs | 3–7 yrs | 7–18 yrs |
|---|---|---|---|---|
| *P. aeruginosa* | 6.45% | 10.57% | 34.44% | 44.08% |
| *S. aureus* | 25.8% | 24.73% | 30.10% | 36.55% |
| MRSA | 4.3% | 5.37% | 9.67% | 7.52% |
| *E. coli* | 11.82% | 12.9% | 4.30% | 10.75% |
| Klebsiella sp | 3.22% | 5.36% | 16.1% | 13.97% |
| *S. maltophilia* | – | – | 2.15% | 3.22% |
| *Aspergillus fumigatus* | 1.07% | 4.30% | 5.37% | 9.67% |
| *Candida albicans* | 15.05% | 5.37% | 11.82% | 24.73% |

## 97 *Clostridium difficile* colitis in transplanted cystic fibrosis patients.

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**Background:** Despite a large carriage rate of *C. difficile* amongst cystic fibrosis (CF) patients, *C. difficile* associated disease (CDAD) is rather rare. In case of lung transplantation, the incidence and clinical aspects of CDAD in this patient population is not well known.
**Aim:** The aim of the study was to compare the incidence and clinical characteristics of CDAD between transplanted and non-transplanted CF subjects.
**Methods:** We reviewed the medical files of all CF patients who presented CDAD from January 1998 to December 2004 and compared the incidence, clinical aspects and outcome between non-transplanted and transplanted patients.
**Results:** Between 1998 and 2004, 106 CF patients were followed at our clinic of whom 15 underwent lung transplantation before and 34 after 1998. The incidence density of CDAD was higher in transplanted CF patients as compared with non-transplanted CF patients (24.2 vs. 9.5 episodes/100000 patient-days; RR: 2.93 [1.41–6.08]; P = 0.0044). Diarrhoea was a very frequent feature, but was notably absent in 20% of the cases. Rates of moderate and severe colitis were similar in both groups. However, only transplanted CF patients developed complicated disease. Two transplant recipients died because of CDAD.
**Conclusion:** CF patients, who undergo lung transplantation, are at a higher risk for developing *C. difficile* colitis and seem to present more often atypical and/or complicated disease. CDAD should be part of differential diagnosis in case of digestive symptoms, even in the absence of diarrhoea, and requires early treatment.

## 98 *Burkholderia pseudomallei* in a cystic fibrosis (CF) patient in Brazil: an emerging pathogen?

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*B. pseudomallei* is a rare isolate from CF patients outside endemic areas. This is the first report of *B. pseudomallei* recovered from the sputum of a CF patient in Brazil.
This is a 17-year-old female with CF and concomitant diabetes mellitus (DM), from Barra dos Bugres, MS, a tropical region of Brazil. Despite the diagnosis of DM and chronic lung infection by *P. aeruginosa* since 1998, her pulmonary disease was well controlled, as shown by normal lung function tests (FEV1 102%) and a chest high resolution computer tomography with only minor bronchiectactic changes (2002). Since 2004 her clinical condition gradually deteriorated, and she presented frequent respiratory exacerbations, a FEV1 drop to 60%, and recurrent radiological signs of infection and atelectasis, along with right upper lobe bronchiectasis. A nonfermentative Gram-negative rod was recovered from her sputum in 2004 but its precise identification was inconclusive. Since 2005, however, *B. pseudomallei* has been repeatedly identified (phenotypic tests confirmed by PCR 16S rRNA gene amplification and sequencing) in her sputum. This organism has only rarely been described in CF patients, most cases being related to travelling to an endemic region. The patient has received many courses of antibiotics with clinical response; eradication, however, was not achieved.
**Conclusion:** Isolation of *B. pseudomallei* from CF patients may be related to clinical deterioration with consequent prognostic and therapeutical implications.

## 99 Sensitivity of recA PCR for direct detection of *Burkholderia cepacia* complex (BCC) from sputum of CF patients

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**Background:** Nucleic acid tests are emerging as practical diagnostic tools for rapid identification and typing of BCC, also enabling direct detection of bacterial DNA from patient's sputum. In this study, sensitivity and diagnostic impact of recA PCR, directly performed on sputum samples were evaluated.
**Methods:** Sputum samples from 28 CF patients were spiked with BCC reference strain with approximate final concentrations of $10^3$ to $10^9$ cfu/ml, having four samples for each concentration. All sputa were processed with sputolysin and nucleic acid extraction was performed by standart phenol-chloroform method. BCC recA gene was amplified using BCR-1 and BCR-2 primers as previously described.
**Results:** BCC DNA was detected in 13 out of 28 (46.4%) samples. BCC concentrations lower than $10^6$ cfu/ml in 12 sputum samples could not be amplified. All samples (4/4) were positive in sputa with $10^9$ cfu/ml BCC, where 3 out of 4 samples were positive in $10^8$ to $10^6$ cfu/ml concentration range.
**Conclusions:** BCC detection limit of recA PCR from sputum was $10^6$ cfu/ml. Diagnostic impact of single-round recA PCR directly from sputum seemed to be limited to chronically-infected patients where bacterial loads of $10^7$–$10^9$ cfu in sputum was noted. The most efficient molecular detection method for BCC directly from patients' sputa need to be determined particularly in the case of newly colonized patients usually expressing lower bacterial counts.