171 *Staphylococcus aureus* with decreased susceptibility to glycopeptides in cystic fibrosis patients

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Cystic fibrosis (CF) patients are frequently colonized or infected with Staphylococcus aureus. Recent reports suggest modifications in isolates epidemiology with strains displaying particular virulence or antibiotic resistance pattern.
We describe the clinical and microbiological features of 2 cases of hetero-Glycopeptide-Intermediate resistance *S. aureus* (h-GISA) isolation in CF patients, one of them having received long-term teicoplanin therapy. The strains were characterized by antibiotyping, population analysis, determination of SCCmec (1 méticillino-resistant strain) and agr types, as well as Sequence Type. They were unrelated to each other. We discuss their epidemiology and pathogenicity.
We currently investigate a collection of 150 strains of *S. aureus* for its susceptibility to glycopeptides. In particular, a new Etest vancomycin-teicoplanin strip for detection of GISA/h-GISA is evaluated. Preliminary results showed that the method failed to detect the two previous h-GISA isolates whereas the reference strain Mu3 was correctly categorized.
S. aureus strains with decreased susceptibility to glycopeptides were previously associated to broncho-pulmonary exacerbation, failure of glycopeptides therapy and clonal spread. Although very rarely reported during CF, these strains may be underestimated in CF patients due to their detection difficulty despite the new tools proposed in routine practice of microbiological laboratories.

172 Characteristics of Staphylococcus aureus small colony variants (SCV) in CF patients

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Background: *S. aureus* SCV are usually associated with persistence in CF airways. This study was aimed to determine the prevalence and antibiotic susceptibilities of *S. aureus* SCV and the demographic characteristics of SCV positive patients.
Methods: Sputum and throat swab specimens of 248 patients were examined during February 2007-January 2008. SCV suspected colonies were confirmed by latex agglutination, coagulase test and S.aureus nucA PCR. Antibiotic susceptibilities were tested by microdilution method. Medical records were reviewed for data collection of SCV positive cases.
Results: A total of 48 SCV were detected in 20 of the 248 patients (8.1%). All the suspected SCV colonies were confirmed as S.aureus. The mean age of SCV isolated patients was 14.4 years. SCV prevalence was significantly higher above age 11. Cocolonization with P.aeruginosa was 55.6% in SCV positive patients whereas it was 28.6% in non-SCV patients ($p < 0.05$). Prior antibiotic use of the SCV positive patients were most frequently beta-lactam beta-lactamase inhibitor combinations (12/19) and ciprofloxacin (12/19). Resistance rates for SCV and their normal S.aureus counterparts were 10.4%, 0% for oxacillin; 6.3%, 0% for gentamicin; 14.6%, 18.2% for ciprofloxacin; 16.7%, 3% for TMP-SXT; respectively. MIC values for tigecycline were higher in SCV isolates. No resistance was detected for vancomycin and linezolid.
Conclusion: Since SCVs are more resistant to antibiotics and have a tendency to resist intracellular killing, it is important to consider and report S.aureus SCVs in CF. Presence of P.aeruginosa in the CF airways might be considered as a predisposing factor for evolution towards *S. aureus* SCV.

173 *Staphylococcus aureus* small colony variants (SASCVs) in CF pts treated at Children's Memorial Health Institute, Warsaw, Poland

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The aim of the study was to determine the frequency of spontaneous and inducible SCV formation in SA isolates obtained from CF pts, to determine antibiotic susceptibility of natural SA (NSA) and SASCV on the basis of minimal inhibitory concentration (MIC) and to determine possible effect of antibiotic treatments in selection of these strains.
Material and Methods: Total of 279 SA strains isolated from 33 CF pts (14 boys, 19 girls, mean age 7.5 years) and 40 isolates from the upper airway from non CF-pts were analysed. The study lasted 8 years. The medical records provided the information about the antibiotic therapy: betalactams 36%, macrolides 21%, fluorochinolones 14%, aminoglycosides 13% (mainly in nebulisation).
Results: SASCV were obtained from 13% isolates from 5 pts. In 3/5 pts SASCV and NSA were isolated simultaneously. In 4/5 pts *P. aeruginosa* (PA) colonisation was found. 4/5 pts received aminoglicosides before the study. SASCV presented decreased susceptibility to some antibiotics in comparison to NSA (MIC SASV vs MIC NSA): gentamicin (32 vs O.75 mg/L), erythromycin (256 vs 0.5 mg/L), ciprofloxacin (8 vs 0.75 mg/L). In the subinhibitory gentamicin concentration SA strains from CF pts formed SCVs more frequently (55%) than isolates from non-CF pts (20%).
Conclusions: The characteristic feature of SA strains, persistently colonizing the CF pts' airways is the formation of SCV. The SASCV formation is affected by the presence of PA. The study has also shown that SASCV formation in CF pts may be associated with inhaled aminoglycoside treatment. SASCV's susceptibility to antibiotics is decreased.

174 Diversity of Candida spp in adult CF patients

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Introduction: Pulmonary infections in Cystic Fibrosis (CF) are predominantly bacterial but the incidence and diversity of fungi as lung pathogens is increasing. Some fungi such as Aspergillus are known to cause significant morbidity in CF but other fungi have not yet been evaluated for their prevalence or clinical impact. This prospective, cross-sectional observational study set out to determine the prevalence and speciation of *Candida spp* in an adult CF cohort.
Methods: Sputum samples were collected from 36 patients attending the Manchester Adult CF Unit. After homogenization with Dithiothreiotol, 10 μL of sputum was inoculated onto sabauroud (SAB) agar and chrome agar then incubated for 72 hours. Yeast colonies were sub-cultured into germ cell tubes for further microscopic identification. Remaining sputum was processed in a molecular laboratory using the MycXtra™ (Myconostica Ltd) fungal DNA extraction kit. Extracted DNA was stored for *Candida* DNA microarray analysis.
Results: SAB agar cultured yeast in 24 of 36 samples (67%). Chrome agar and germ cell culture identified >1 *Candida* species in 7 of the 24 positive samples. 4 plates had species that were unidentifiable. Of those colonies identified the following species were found: *Candida albicans/dublinensis* 22, *Candida glabrata* 5, *Candida tropicalis* 1, *Candida parapsilosis* 1. These species are currently being tested for antifungal resistance. As there are unidentified species, we are currently performing molecular identification with a *Candida* DNA microarray.
Conclusion: There is a wide diversity of *Candida spp* in adult CF sputum. Fungal-bacterial interactions are important. More research is required into the clinical relevance of these yeasts.