

**Conclusions:** This is the first description of PA CL-resistance in Italian CF patients. Although in very low percentage (2.2 %) this emergence needs concern in the extensive and prolonged use of this important therapeutic option. Moreover, in the CF microbiology workout, seems to be mandatory the CL (or polymixin B) routinely testing in all PA isolates.

**[P25\*] PSEUDOMONAS AERUGINOSA (PA) FROM CONTAMINATED SWIMMING POOLS: A SOURCE OF ACQUISITION FOR CYSTIC FIBROSIS (CF) PATIENTS?**

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**Background:** The initial source of PA acquisition remains unknown in most CF patients (pts), but the environment is one of the possible sources. Water is the natural habitat for PA, but at the moment, there are limited published data about the prevalence of PA in swimming pools and the real risk of acquisition for CF pts. **Aim:** Characterize PA recovered from swimming pools, in order to determine genetically similarity with strains isolated from CF pts.

**Methods:** A collection of 30 strains from 7 swimming pools of Ligurian region, isolated by the ARPAL Microbiology Laboratory, were characterized for antibiotic resistance and hypermutable (HMP) phenotype by disk-diffusion and E-test (Macia et al 2004). All strains were genotyped by Box-PCR (Rademaker J.L.W, et al 1998) and the genetically relatedness was assessed by Gel Compare II analysis. A comparison with 240 previously genetically characterized strains isolated from 121 pts of Genova CF Centre was performed.

**Results:** All the strains were susceptible to Ciprofloxacin, Tobramycin and Colistin, the antibiotic resistance (%) was: Meropenem 20, Gentamicin 7, Ceftazidim 3, Piperacillin/Tazobactam 3, Ticarcillin/Clavulanate 3, Amikacin 3, Ticarcillin 3 and Aztreonam 3. 1/30 (3%) strain showed a HMP phenotype. By Box-PCR a total of 16 genotypes were found: 11 genotypes were not shared, while 5 were shared. In particular 5 clusters were found, with different size of swimming pools: 3 of 2 and 2 of 3. No genetic correlation was observed between swimming pools and CF strains.

**Conclusions:** This study shows that PA recovered from swimming pools have a phenotype with low resistance to antimicrobial agents and a low occurrence of HMP strains. Our results show that a large number of different PA genotypes are present in swimming pools of Ligurian region. Although our data did not demonstrate any PA genotype shared by CF pts and swimming pools strains, care should be taken anyway suggesting CF pts to attend any swimming pool.

\*This Poster has been presented also as Oral Communication, by invitation of the Scientific Committee

**[P26] PERSISTENCE OF ACHROMOBACTER XYLOSOXIDANS IN CF PATIENTS: RESULTS OF A MOLECULAR STUDY**

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**Introduction:** *Achromobacter xylosoxidans* (*A. xylosoxidans*) has been increasingly recognized as a cause of respiratory tract colonization in Cystic Fibrosis (CF). The pathogenic role of this Gram-negative bacteria is yet unclear. The present study was conducted to investigate the epidemiology of *A. xylosoxidans* isolates recovered from CF patients. Misidentification of *A. xylosoxidans* can occur, therefore the presence of respiratory tract infections caused by this bacteria might be underestimated. Epidemiology of *A. xylosoxidans* in CF is not well understood: it can cause temporary or persistent infection in the respiratory tract. A total of 55 isolates were detected persistently in 8 out of 260 CF patients over a prolonged period of time (from 2 to 8 years).

**Aims:** Sequential *A. xylosoxidans* strains of 8 CF patients were genotyped in order to detect if these patients are persistently colonized, or whether a cycle of acquisition and clearing has occurred.

**Methods:** Lactose-negative on McConkey agar and oxidase-positive colonies were firstly identified using API20NE (bioMérieux), then confirmed by PCR. This molecular method allows to identify *A. xylosoxidans* recovered from CF patients sputum with a sensitivity of 100% and specificity of 97%, in order to distinguish *A. xylosoxidans* isolates from several related glucose non-fermenting species. Bacterial isolates were fingerprinted by RAPD-PCR.

**Results:** Three out of 55 (5.5%) isolates were incorrectly identified as *A. xylosoxidans* and were not included in the epidemiological study. Isolates collected from 5 out of 8 patients exhibited identical RAPD profiles, indicating that the same strain persisted in the lungs of these patients. The remaining three patients are alternatively colonized by few (2 or 3) different genotypes.

**Conclusions:** In an attempt to determine whether CF patients acquired a single isolate that they kept for years, or were periodically recolonized, serial isolates on a subpopulation of 8 patients were examined. It appears that the majority of patients (62.5%) have unique isolates, and only few patients are colonized by more than one genotypes. A better understanding of the epidemiology of *A. xylosoxidans* may

help us to clarify the pathogenic potential of this unusual Gram-negative bacteria in CF lung disease.

**[P27] PSEUDOMONAS AERUGINOSA PRODUCING METALLO-β-LACTAMASES IMP-13 IN A PATIENT WITH CYSTIC FIBROSIS**

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**Introduction:** *Pseudomonas aeruginosa* is the most important opportunistic pathogen in Cystic Fibrosis (CF) patients. The presence in *P. aeruginosa* of acquired metallo-β-lactamase (MBL), that confers broad-spectrum resistance to β-Lactam antibiotics, represents an alarming phenomenon in clinical practice. Acquired resistance determinants in patients with CF are not commonly found and *bla*<sub>IMP</sub> genes have never been described. In this study we report about the first identification of an IMP-type MBL in isolates of *P. aeruginosa* from a CF patient.

**Methods:** Isolates of *P. aeruginosa* with a profile suggesting the production of MBL collected from eight patients attending the Ospedale Bambin Gesù in Rome have been analysed. Hybridization, PCR and DNA sequencing have been used to detect *bla*<sub>IMP</sub> genes. Retrospective isolates have been included in the study. Clonality of strains producing MBL have been analysed by genotyping work (RAPD/PFGE).

**Results:** From one out of eight patients (an 8-year-old girl), a *P. aeruginosa* strain producing an IMP-type resistance determinant, encoded by the gene variant *bla*<sub>IMP</sub> has been isolated. The analysis of the retrospective isolates from the patient, as well as their clonality, demonstrated the persistence of the isolate producing IMP-13 along a period of 3 years. Besides, the analysis has highlighted the presence of this resistance determinant since the first time the patient has been colonized by *P. aeruginosa*. The clinical course of the patient has been characterised by pulmonary exacerbations, with a slow but progressive decline of pulmonary function (FEV<sub>1</sub>: 88%) despite repeated antibiotic cycles (aminoglycosides and quinolones).

**Conclusions:** Metallo-β-lactamase (MBL) *bla*<sub>IMP</sub>, widespread in Italy, has been isolated for the first time in *P. aeruginosa* strains from a patient with CF. This is an alarming phenomenon which can have important implications for the therapeutic as well as the surveillance field.

**[P28] "ANGELA'S CASE"**

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**Background:** We would like to introduce the clinical case of "Angela", an Albanian girl aged 24 months, who has been diagnosed by neonatal screening at our CF Centre (Meyer Hospital, Florence, Italy).

Following and taking care of A. and her family represents and has represented since CF diagnosis moment a very difficult engagement because of the many unsuccessful attempts of creation of a good collaborative relationship with A.'s parents. Our difficulty is mainly correlated to the mother's parental and attachment style that has not permitted to understand for certain how she takes care of A. and manages her daughter's illness.

We hypothesize that the mother needs to consider the symptoms of her child as CF related even if there is no clinical evidence. This hypothesis originated from the different communications that the mother usually gives to the various operators of the CF Centre about what happens to A. and how she faces the different illness aspects and moments; doing that she shows an ambivalent relational style: frequent treatment requests accompanying the difficulty or incapacity to follow the medical indications, for instance she requested invasive medical checks for A. because of abdominal-ache and at the same time she gave A. coke or other drinks with gas; she did not confess that frankly but she minimized what happened when we asked information about that. Moreover we have the perception of high risk for A.'s physical and psychological health: she keeps facing many hospitalizations because of ruinous situations of frequent vomiting and loss of weight that turn out well with adequate therapy and controlled diet in a few days in hospital.

**Clinical situation and clinical course:** pancreatic insufficiency; frequently hospitalization for vomit, food refuse and very important weight loss; pulmonary disease without main infection exacerbations, not *Pseudomonas aeruginosa* colonisation. During last hospitalization, for recurrent vomit and weight loss, we tried to perform:

- Esophageal pH 24 hours monitoring: not concluded because A. take off the tube;
- Gastrointestinal contrast radiographic study on narcotic state;
- Endoscopy, and ultrasound only showed a medium gastroesophageal reflux;
- Physiotherapy programme; feeding programme; pancreatic and antacid therapy, family psychological support.

**Discussion:** We ask ourselves how and how much it is necessary to protect A. from her mother's nurturing style by more important interventions to control the mother's psychological capacity about her role in the relationship with her daughter and in the management of the illness.