191* Clinical value of *Aspergillus* detection in sputum obtained from 84 patients with cystic fibrosis

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Aspergillus are filamentous fungi which colonize the respiratory tract of patients with cystic fibrosis (CF). As a consequence of colonization, sensitization to *Aspergillus* can induce allergic bronchopulmonary aspergillosis (ABPA). The objective of this study was to evaluate the clinical value of *Aspergillus* detection in the management of patients with CF.

208 sputum obtained from 84 patients with CF from the CF Centers of Rennes Brittany, France, were analyzed during a 6 month-prospective study. *Aspergillus* detection was performed both by mycological culture and by quantitative PCR using 2 sets of primers targeting the rRNA 5.8s and *A. fumigatus* mitochondrial RNA.

Among patients without ABPA, 51% were colonized by *A. fumigatus*, either by classic mycological culture or by PCR. Beside, 50% of patients with classic ABPA diagnostic criteria (clinical findings, immediate hypersensitivity skin test for *A. fumigatus*, total and *A. fumigatus* specific serum IgE, anti-*Aspergillus* precipitins, eosinophil count) showed a positive detection of *Aspergillus* positive. The correlation between mycological culture and quantitative PCR reached 91.8%. A lower sensitivity of mycological culture was observed in patients with ABPA receiving an antifungal treatment. *Aspergillus* detection in the sputum of patients suffering from CF without ABPA is a useful and non-invasive tool for the early characterization of patients at risk for sensitization. During ABPA, *Aspergillus* detection is an obvious marker of antifungal treatment failure when it is instituted and must incite practitioner to investigate several causes of failure.

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192 Tolerability of voriconazole in children with cystic fibrosis and allergic bronchopulmonary aspergillosis (ABPA)

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Introduction: Allergic Bronchopulmonary Aspergillosis (ABPA) is a hypersensitivity reaction to *Aspergillus fumigatus* and a complication of CF, the treatment of which is based on the use of systemic steroids. In patients with serious complications from corticosteroids or in refractory to corticosteroids ABPA, the use of antifungal agents contributes to the discontinuation or the reduction of steroids.

Purpose: To describe the tolerability of voriconazole as well as its results in the treatment of ABPA in CF patients.

Material and Methods: In our center, we treated 10 ABPA patients aged between 5–20 years old (mean age: 14 years, males: 7, females:3) with voriconazole (dose: 8 mg/kg/24 h, maximum: 400 mg) for a period of >6 months.

Results: 5 of our children presented photosensitivity/exanthema, 2 of them presented also exfoliative dermatitis. 5 of the patients presented elevation of the hepatic enzymes, mainly of γ-gt. One of them with serious elevation of the hepatic enzymes presented improvement of the hepatic function after diminishing the dose of voriconazole and when we restarted the full-dose of the antifungal agent the aforementioned side effect did not reappear. One of the patients presented disturbances of vision (bright spots) and another headaches at the beginning of the treatment.

Conclusions: The use of voriconazole was safe, and was associated with transient side effects in 6 out of the 10 children.

193 Analysis of fungal–bacterial community interactions in cystic fibrosis airway secretions

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The airways of cystic fibrosis patients are susceptible to colonisation by fungal pathogens. The most significant of these is *Aspergillus fumigatus*, which can lead to the development of allergic bronchopulmonary aspergillosis. Previous studies have explored prevalence and impact on patient health of both *A. fumigatus* and other fungal species known to colonise CF sputum, such as *Candida albicans*.

However, to date, such studies of fungal species in isolation have failed to address an important factor. The CF airway is typically colonised by a complex array of bacterial species. This bacterial community can profoundly influence, and be influenced by, the presence of fungal species. For example, pseudomonads are known to be capable of producing a wide range of compounds that influence the behaviour of fungal species, both positively and negatively. Increasingly, evidence is emerging that suggests chemical signalling pathways may exist between fungi and bacteria.

In this study we used molecular profiling techniques to determine both the bacterial and fungal composition of sputum samples from 14 adult CF patients. T-RFLP, based on the 16S and 18S ribosomal RNA genes, was used to profile the bacterial and fungal communities respectively. Initial data suggests fungal pathogens are particularly prevalent in patients with low diversity bacterial populations. Further, there is a marked separation between detection of fungal pathogens in the CF airways, which is common, and the presence of significantly large colonisations, which is relatively rare. These data highlight the possibility of important fungal–bacterial interactions in vivo and underline the need for further investigation in this area.

194 Emergence and spread of a phylogenetic cluster of Corynebacteria in cystic fibrosis patients

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We report in this study 12 cases of Cystic Fibrosis (CF) patients eventually colonized or infected with Corynebacterium pseudodiphteriticum/propinquum in our reference centre for children.

The sputa were collected from children and adults CF reference center at Marseille (France). Sixteen Corynebacteria strains were isolated from 12 children sputum samples and none from adults. Among twelve patients, seven were associated with cough. All the strains were identified by phenotypic methods (Api Coryne database system 2.0) and by 16SrRNA and partial rpoB gene sequencing. Fifteen Corynebacteria belong to the Corynebacterium pseudodiphtericum/propiquum cluster with a confidence level >98% whereas one strain could not be identified correctly by gene sequencing and may represent a new Corynebacterium species.

Corynebacterium were never reported in CF patients probably because these bacteria are usually considered as contaminant of culture of sputum samples. Furthermore different closely related Corynebacteria were isolated during time in the same patient suggesting a mixed colonization and/or infection. The significance of the isolation of these bacteria known to cause various pulmonary diseases and being a part of the normal microflora of the higher respiratory tract remains unclear but may contribute to the progression of the disease. Epidemiological and clinical studies are warranted to define the role of these bacteria in the progression of CF disease.

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