

Serologic IgE Immune Responses Against *Aspergillus fumigatus* and *Candida albicans* in Patients With Cystic Fibrosis*

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Objectives: The objectives of this study were to determine the prevalence of *Aspergillus fumigatus* and *Candida albicans* in the sputa of patients with cystic fibrosis (CF), to assess serologic IgE responses of these patients to the presence of fungi in the sputum, to evaluate what effect this may have on clinical status, and to determine how the above-mentioned factors relate to allergic bronchopulmonary aspergillosis (ABPA).

Patients: Seventy-six CF patients (40 male and 36 female patients; age, 15.3 ± 8.7 years [mean $\pm$ SD]) were studied.

Measurements and results: A total of 1,239 sputum samples from 66 patients were cultured for fungi. *A fumigatus* was grown in 256 sputum specimens (20.7%), and *C albicans* was grown in 588 sputum samples (47.5%). Forty patients (60.6%) had at least one positive culture finding for *A fumigatus*, and 58 patients (87.9%) had at least one positive culture finding for *C albicans*. Forty-nine patients (64.5%) were sensitized to *A fumigatus*, and 20 patients (26.7%) were sensitized to *C albicans*. No correlation was found between the finding of *A fumigatus* in sputum and IgE to *A fumigatus*. Only patients who had at least one positive culture finding for *C albicans* had IgE to *C albicans* develop. Lung function values and chest radiograph scores were not significantly lower in patients sensitized to either *A fumigatus* or *C albicans* as compared to nonsensitized patients. Of the 20 patients sensitized to *C albicans*, 10 patients had confirmed ABPA and 10 patients had some immunologic characteristics of ABPA.

Conclusions: A high prevalence of colonization and sensitization to *A fumigatus* and *C albicans* in CF patients was observed. The sensitization to these fungi was not related to the clinical severity. IgE to *C albicans* may be an immunologic marker related to the development of ABPA in patients with CF. (CHEST 2002; 121:782–788)

Key words: allergic bronchopulmonary aspergillosis; *Aspergillus fumigatus*; *Candida albicans*; cystic fibrosis; IgE

Abbreviations: ABPA = allergic bronchopulmonary aspergillosis; CF = cystic fibrosis; PBS = phosphate-buffered saline solution

Fungal colonization of the airways is frequently observed in patients with cystic fibrosis (CF).^{1,2} The fungi most frequently recovered from respiratory secretions are *Aspergillus fumigatus* and *Candida albicans*.^{3,4} The colonization of the respiratory

tract by *A fumigatus* usually provokes IgE reactivity to this fungus in patients with CF, although not necessarily manifesting as allergic bronchopulmonary aspergillosis (ABPA).^{5,6} ABPA occurs with a prevalence rate of 0.5 to 15% in CF patients.^{3,7} The increased probability of the development of specific IgE to *A fumigatus* in CF patients is probably due to the ability of this fungus to colonize the respiratory tract.⁵ Although the contributory role of fungi in causing lung damage in patients with CF is not well

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defined, some investigators^{8,9} suggest that the sensitization to *A fumigatus* *per se* contributes to the disease process.

Currently, there are no data indicating that *Candida* sp play a role in CF lung disease. The presence of specific IgE antibodies to *C albicans* has been reported in ABPA subjects,¹⁰ meaning that the role of this yeast in ABPA merits consideration.

The aims of this study were to determine the prevalence of *A fumigatus* and *C albicans* in the sputa of CF patients, and to assess IgE responses of these patients to the presence of fungi in the sputum. We also evaluated the effect of sensitization to *A fumigatus* and *C albicans* on clinical status, total serum IgE, and eosinophils in peripheral blood. Finally, we determined how the above-mentioned factors relate to ABPA in patients with CF.

MATERIALS AND METHODS

Patients

Seventy-six patients with CF (40 male and 36 female patients; mean[$\pm$ SD] age, 15.3 $\pm$ 8.7 years; range, 1 to 36 years) recruited before May 1992 were prospectively studied during their routine clinic visits. All patients were studied at Ramón y Cajal Hospital in Madrid; 72 patients were recruited from the CF Unit at this hospital and 4 patients were referred from Virgen del Rocío Hospital in Seville, Spain. In all CF patients, the diagnosis was established from sweat chloride concentrations > 60 mEq/L on at least two specimens measured by quantitative iontophoresis. Informed consent was obtained from all participants or their guardians, and the study was approved by the local ethical committee.

Study Protocol

Patients were prospectively evaluated every 3 to 4 months over a period ranging from 2 to 6 years, between May 1992 and May 1998. Additional visits and analyses were performed whenever considered necessary by the clinician. During the 6 years of the study, all respiratory secretions were cultured for fungi. During the last 3 to 4 years of the study, at least one blood sample was analyzed every 8 months in each patient. Routine assessment was done once yearly, including chest radiography (scored by the Brasfield system),¹¹ lung function testing, and immediate skin testing to *A fumigatus*. Analyses included total serum IgE (478 samples), IgE to *A fumigatus* (471 samples) and IgE to *C albicans* (423 samples), precipitins against *A fumigatus*, and peripheral blood eosinophil count. Skin-prick tests to other common aeroallergens were performed once during the study. Patients with less than four sputum samples in the last year of the study were eliminated from the study in those analyses that included sputum cultures; otherwise, their data were used for other measurements.

Definition of Specific Diagnosis

Several groups were defined according to the microbiology results during the study: patients who had at least one positive sputum culture finding for *A fumigatus* or *C albicans* (*A fumigatus*-recovered or *C albicans*-recovered groups, respectively), and

patients who never had *A fumigatus* or *C albicans* in their respiratory secretion cultures at any time (*A fumigatus*-free or *C albicans*-free groups, respectively). Sensitization to *A fumigatus* and/or *C albicans* was considered to be present when specific IgE titer (Pharmacia CAP System; Pharmacia Diagnostics; Uppsala, Sweden) to *A fumigatus* and/or *C albicans* was positive (> 0.35 kilounits/L) at a given visit. Atopy was defined as the presence of at least one positive immediate skin test response to commercial extracts of common aeroallergens (not including *A fumigatus* and *C albicans*).

The diagnosis of ABPA was based on a minimum of six of seven criteria as described by Nelson et al.⁷ These criteria included episodic bronchial obstruction (asthma), positive sputum culture finding for *A fumigatus*, positive immediate skin test response to *A fumigatus*, increased total serum IgE concentrations (IgE > 400 kilounits/L), specific serum IgE antibodies to *A fumigatus*, presence of serum precipitins to *A fumigatus*, and a history of pulmonary infiltrates. None of the CF patients received antifungal treatment during the study. Although detection of *A fumigatus* in sputum by smear or culture is considered a minor criterion for the diagnosis of ABPA,³ lack of recovery of *A fumigatus* from sputum does not rule out the diagnosis of this entity.¹²

Total and Specific IgE Determinations

Measurements of total serum IgE and specific serum IgE concentrations to *A fumigatus* were performed in all patients. Specific serum IgE concentrations to *C albicans* were performed in 75 patients. Total serum IgE (kilounits per liter) was measured by Pharmacia CAP System IgE fluoroenzyme immunoassay (Pharmacia Diagnostics). Total serum IgE was considered elevated when the IgE concentrations were higher than the mean $+ 1.96$ SD of expected values according to age. The serum concentrations of specific IgE antibodies against *A fumigatus* and *C albicans* were measured by the Pharmacia CAP System radioallergent test fluoroenzyme-immunoassay (Pharmacia Diagnostics). Pharmacia CAP System scores ≥ 0.35 kilounits/L were regarded as positive, as recommended by the manufacturer.

Specific IgE Inhibition Assays

In order to investigate the specificity of the Pharmacia CAP System technique, competitive IgE inhibition assays with the sera of six patients with Pharmacia CAP System results to *C albicans* ≥ 3.5 kilounits/L were performed. *C albicans* extract was used in the solid phase, and either *C albicans* or *A fumigatus* were used as inhibitor allergens. Volumes of 20 μ L in four progressive 10-fold dilutions of the inhibitor extract in phosphate-buffered saline solution (PBS), and 20 μ L of PBS as negative control were preincubated with 50 μ L of serum for 30 min at room temperature. Thereafter, the assay was continued following the standard Pharmacia CAP System technique. Results were expressed as the percent inhibition of Pharmacia CAP System obtained with the inhibitor allergen as compared with a control assay with PBS inhibitor.

Precipitins Against *A fumigatus*: Serum precipitating antibodies to *A fumigatus* antigens were measured by means of the double immunodiffusion technique with a panel of standard antigens (Pasteur Diagnostic; Paris, France). A visual line of precipitation between serum samples and antigen constituted a positive determination.

Peripheral Blood Eosinophils: Total peripheral blood eosinophil count was determined in all the blood samples.

Skin Tests: Skin tests to *A fumigatus* were performed by the prick method in 66 patients. Skin-prick tests were also performed with a battery of common inhalant allergens in 59 patients. The

following allergenic extracts were used: *A fumigatus* 5% weight/volume (Stallergènes; Antony, France), and a battery of commercially available common inhalant allergens, including *Dermatophagoides pteronyssinus*, *Dermatophagoides farinae*, grass and olive tree pollen, *Cladosporium herbarum*, *Alternaria alternata*, *Penicillium notatum*, and dog and cat epithelium (ALK-Abelló; Madrid, Spain). Histamine chlorhydrate (10 mg/mL) and normal saline solution (0.9%) were used as positive and negative controls, respectively. All skin-prick test results were read after 15 min and were considered positive if the wheal size was ≥ 3 mm in the presence of a negative reaction to the saline solution control. Intradermal tests to *A fumigatus* aqueous extract 1% weight/volume (ALK-Abelló) were performed only in those patients who had a negative skin-prick test response to these fungal extracts. Immediate skin test reactivity to *A fumigatus* was considered positive by either a positive epicutaneous or intradermal test response.

Sputum Processing

Respiratory secretions (sputum samples if obtained, or post-tussive deep pharyngeal swabs) were recovered at each clinic visit and cultured for fungi, with a maximum of one sample a month. The samples were cultured on Sabouraud-chloramphenicol and Sabouraud-chloramphenicol-actidione with antibiotics to inhibit the bacterial overgrowth. The culture plates were cultivated at two temperatures (30°C and 37°C). Cultures were evaluated for growth at 24 h, 7 days, and 4 weeks later.

Lung Function Testing

Lung function tests were carried out on 60 patients > 5 years old who were able to perform a valid spirometry. FEV₁ and FVC were measured without bronchodilation on a Vmax 20 Spirometer (SensorMedics Corporation; Yorba Linda, CA). Results were expressed as a percentage of predicted values for age, sex, race, weight, and height based on the standards of the European Community for Coal and Steel.¹³

Statistical Analysis

A comparison of the different groups was determined with the Pearson χ^2 test with Yates correction for categorical data and the Mann-Whitney *U* test for comparison of rank data. The Spearman correlation test was used for comparison of rank data with quantitative data. The Bartlett test was used to study the homogeneity of variance. Categorical data with quantitative data were compared with the Student's *t* test if the variances were homogenous, and with the Mann-Whitney *U* test if the variances were not homogenous. Correlation coefficients were determined using Pearson linear regression analysis. A multivariable analysis was used to study possible associations between sensitization to *A fumigatus* and *C albicans*, age, gender, total IgE, total eosinophil count, and lung disease defined by spirometry and the Brasfield score. Differences associated with probabilities < 0.05 were considered significant using a two-tailed *p* value. All analyses were done with statistical software (DBase-IV, Epiinfo version 6.04-b; Borland International; Scotts Valley, CA; and SPSS version 8.0; SPSS; Chicago, IL).

RESULTS

During the study, 76 CF patients were followed up as outpatients. The mean Brasfield score for all

CF patients was 18.5. The mean FVC (percentage of predicted value) was $79.85 \pm 26.24\%$, and FEV₁ (percentage of predicted value) was $72.09 \pm 31.28\%$. During these 6 years, 1,239 respiratory secretion samples from 66 patients were sent for yeast culture (mean, 18.8 cultures per patient; range, 4 to 48). Two hundred fifty-six respiratory secretions had *A fumigatus* (20.7%) and 588 secretions had *C albicans* (47.5%). Forty patients (prevalence of 60.6%) had at least one positive culture finding for *A fumigatus* (*A fumigatus*-recovered group), and 58 patients (prevalence of 87.9%) had at least one positive culture finding for *C albicans* (*C albicans*-recovered group). Thirty-nine patients (51.3%) had elevated total serum IgE.

There was no difference in either the total IgE or specific IgE concentrations to *A fumigatus* between the *A fumigatus*-free group and the *A fumigatus*-recovered group. In addition, no correlation was found between the presence of *A fumigatus* in sputum and a positive IgE titer to *A fumigatus*. However, although the total IgE and specific IgE concentrations to *C albicans* showed no significant differences between the *C albicans*-free group and the *C albicans*-recovered group, only patients belonging to the *C albicans*-recovered group had a positive IgE titer to *C albicans* (20 of 58 patients), whereas none of the patients of the *C albicans*-free group had a positive IgE to *C albicans* (0 of 8 patients; *p* < 0.001).

Forty-nine patients (64.5%) were sensitized to *A fumigatus* as determined by a positive IgE titer to *A fumigatus* (> 0.35 kilounits/L). The characteristics of sensitized and nonsensitized patients to *A fumigatus* are shown in Table 1. Lung function values and Brasfield scores were lower in *A fumigatus*-sensitized patients, but the differences were not statistically significant. Concentrations of total serum IgE and peripheral blood eosinophil count were significantly higher in *A fumigatus*-sensitized patients than in those without sensitization to *A fumigatus*. Twenty of 75 patients (26.7%) were sensitized to *C albicans*. All the patients sensitized to *C albicans* showed a concomitant sensitization to *A fumigatus* (*p* < 0.001). Twenty-eight patients were nonsensitized to both *C albicans* and *A fumigatus*. Table 2 shows the characteristics of patients sensitized and nonsensitized to *C albicans*. Lung function values and the Brasfield scores were no different between these two study groups. Total IgE concentrations and peripheral blood eosinophil counts were significantly higher in the *C albicans*-sensitized group than in the nonsensitized group.

IgE reactivity to *C albicans* was almost completely inhibited (90% maximum inhibition) with *C albicans* extract in each of six individual serum with high IgE

Table 1—Characteristics of CF Patients With and Without Sensitization to *A fumigatus**

Characteristics	<i>A fumigatus</i> -Sensitized Patients (<i>A fumigatus</i> IgE $\geq$ 0.35 kilounits/L) [n = 49]	<i>A fumigatus</i> -Nonsensitized Patients (<i>A fumigatus</i> IgE < 0.35 kilounits/L) [n = 27]	p Value
Age, yr	15.77 $\pm$ 7.88	14.70 $\pm$ 10.16	0.58
Male/female gender, No.	22/27	18/9	0.11
Brasfield score	17.34 $\pm$ 4.78	19.26 $\pm$ 4.87	0.10
FVC, % predicted	76.58 $\pm$ 27.69	86.91 $\pm$ 21.81	0.16
FEV ₁ , % predicted	66.96 $\pm$ 31.20	83.18 $\pm$ 29.25	0.07
Total serum IgE, kilounits/L	653.22 $\pm$ 1,039.38	38.30 $\pm$ 40.83	< 0.001
Eosinophils, cells/ μ L	298.31 $\pm$ 188.87	210.76 $\pm$ 135.59	< 0.05

*Data are presented as mean $\pm$ SD unless otherwise indicated.

titers to *C albicans* ($\geq$ 3.5 kilounits/L), whereas it was not significantly inhibited with *A fumigatus* extract, indicating the specificity of this assay.

Evidence of more advanced lung disease by Brasfield score but not by spirometric parameters was associated with increasing specific IgE concentrations to *A fumigatus* ($r = -0.28$; $p < 0.05$) and increasing IgE to *C albicans* ($r = -0.23$; $p < 0.05$). Age was not associated with increased IgE concentrations to *A fumigatus* or with IgE concentrations to *C albicans*. In *A fumigatus*-sensitized patients, we found a significant linear correlation between IgE to *A fumigatus* and total serum IgE concentrations ($r = 0.81$; $p < 0.001$) and between IgE to *A fumigatus* and total eosinophil count ($r = 0.45$; $p < 0.001$). In patients sensitized to *C albicans*, a similar correlation was found between IgE to *C albicans* and total IgE ($r = 0.87$; $p < 0.001$) and between IgE to *C albicans* and total eosinophil count ($r = 0.41$; $p < 0.001$). In addition, there was a close correlation between IgE to *A fumigatus* and IgE to *C albicans* ($r = 0.64$; $p < 0.001$).

In a linear multiple regression analysis in which FEV₁ was the dependent variable and age, gender, total IgE, total eosinophil count, and sensitization to *A fumigatus* and to *C albicans* were covariates, age

alone accounted for almost all the increased risk for a low FEV₁ ($\beta = -0.29$; SE = 0.5; $p = 0.02$). Similar results were observed when FVC was used as a dependent variable. By logistic regression analysis, the sensitization to *A fumigatus* and to *C albicans* did not independently increase the risk of more severe lung disease, defined for the purpose of the analysis as a Brasfield score < 18. Increasing age was closely associated with poor Brasfield score ($\beta = -0.09$; SE = 0.03; $p < 0.001$).

Skin testing in 34 of the 66 patients (51.5%) with *A fumigatus* showed a positive response. Twenty-five patients had a positive skin-prick test response to *A fumigatus*, and 9 patients had a positive intradermal test response to *A fumigatus* with a negative skin-prick test response. No negative conversion was observed over the study period. Skin-prick tests to common aeroallergens were performed in 59 patients, and 35 patients (59.3%) were classified as atopic. Of the 35 atopic patients, all but 6 had a positive skin test response to *A fumigatus*. All patients sensitized to *C albicans* were atopic (positive skin test response to grass pollen and/or animal dander) [$p < 0.01$].

The clinical data for ABPA are summarized in Table 3. Mean age, sex, and pulmonary status of the

Table 2—Characteristics of CF Patients With and Without Sensitization to *C albicans**

Characteristics	<i>C albicans</i> -Sensitized Patients (<i>C albicans</i> IgE $\geq$ 0.35 kilounits/L) [n = 20]	<i>C albicans</i> -Nonsensitized Patients (<i>C albicans</i> IgE < 0.35 kilounits/L) [n = 55]	p Value
Age, yr	14.65 $\pm$ 7.62	15.47 $\pm$ 9.08	0.72
Male/female gender, No.	9/11	30/25	0.64
Brasfield score	16.50 $\pm$ 3.98	18.54 $\pm$ 5.08	0.14
FVC, % predicted	79.93 $\pm$ 26.40	81.57 $\pm$ 26.41	0.84
FEV ₁ , % predicted	72.56 $\pm$ 32.73	74.66 $\pm$ 24.06	0.82
Total serum IgE, kilounits/L	1,288.85 $\pm$ 1,362.47	120.02 $\pm$ 185.96	< 0.001
Eosinophils, cells/ μ L	383.95 $\pm$ 224.79	224.53 $\pm$ 127.31	< 0.05

*Data are presented as mean $\pm$ SD unless otherwise indicated.

Table 3—Comparison Between CF Patients With ABPA and Without ABPA*

Variables	ABPA Patients (n = 10)	Non-ABPA Patients (n = 66)	p Value RR (95% CI)
Age, yr	11.90 ± 9.29	15.92 ± 8.56	0.17
Male/female gender, No.	4/6	36/30	0.50
Brasfield score	17.75 ± 3.77	18.09 ± 5.02	0.85
FVC, % predicted	83.65 ± 28.98	79.27 ± 26.05	0.66
FEV ₁ , % predicted	81.74 ± 16.56	70.61 ± 32.83	0.89
Total IgE, kilounits/L	1,786.40 ± 1,697.55	229.97 ± 420.78	< 0.001
Eosinophils, cells/μL	483.9 ± 252.82	231.25 ± 131.37	< 0.001
IgE to <i>A fumigatus</i> , kilounits/L	34.45 ± 23.35	8.68 ± 17.12	< 0.001
Sensitized/nonsensitized patients to <i>A fumigatus</i> , No.	10/0	39/27	1.26 (1.09 to 1.45) < 0.05
IgE to <i>C albicans</i> , kilounits/L	5.05 ± 8.20	1.35 ± 1.26	0.25
Sensitized/nonsensitized patients to <i>C albicans</i> , No.	10/0	10/55	2.00 (1.29 to 3.10) < 0.001

*Data are presented as mean ± SD unless otherwise indicated. RR = relative risk; CI = confidence interval.

subjects with CF who had ABPA did not differ significantly from the non-ABPA CF patients. ABPA patients were treated with oral corticosteroids, usually starting at 1 mg/kg/d for 2 weeks followed by decreasing dosages for approximately 3 to 6 months.

Mean total serum IgE concentrations and total eosinophil counts were significantly raised in ABPA patients as compared with non-ABPA patients ($p < 0.001$). Moreover, ABPA patients had significantly higher IgE concentrations to *A fumigatus* than the non-ABPA patients ($p < 0.001$). Although the serum IgE concentrations to *C albicans* showed no significant differences between these two groups, all 10 ABPA patients were sensitized to *C albicans* ($p < 0.001$). Each of the 10 non-ABPA patients sensitized to *C albicans* fulfilled the immunologic criteria of ABPA but lacked pulmonary infiltrates or, if these infiltrates were present, responded satisfactorily to antibiotic therapy.

There was a highly significant linear correlation between total IgE and IgE to *A fumigatus* in non-ABPA patients ($r = 0.76$; $p < 0.001$) as well as in ABPA patients ($r = 0.83$; $p < 0.05$). There was also a significant linear correlation between total IgE and IgE to *C albicans* in non-ABPA patients ($r = 0.75$; $p < 0.001$) and in ABPA patients ($r = 0.92$; $p < 0.001$).

DISCUSSION

In our CF population, the prevalence of *A fumigatus* and *C albicans* in respiratory secretions was 20.7% and 47.5%, respectively. Prevalence rates of *A fumigatus* and *C albicans* recovered from the lower respiratory tract secretions seen in the present study are comparable to the prevalence of this fungi in CF patients reported by other authors.^{4,7,14} It has

been suggested that different antibiotic regimens in CF patients may lead to different degrees of colonization, germination of fungal spores, and sensitization. However, the incidence of fungal colonization varies widely between different geographic regions for no obvious reasons.

The ability of *A fumigatus* to grow in the respiratory tract makes exposure to this fungus the most likely explanation for the high incidence of specific immune responses to *A fumigatus* in CF patients.⁵ The humoral immunologic response to *A fumigatus* in CF patients shows tremendous variability, diminishing or disappearing spontaneously.¹⁵ Our results demonstrate a highly prevalent IgE response to *A fumigatus* in accordance with those of El-Dahr et al.⁵

In our study, no correlation was found between the presence of *A fumigatus* in sputum and specific IgE immunoresponses, in agreement with the study of El-Dahr et al.⁵ Nevertheless, we noted that specific IgE response to *C albicans* was always correlated with the presence of this yeast in the respiratory secretions. This might be due to the fact that, while positive sputum culture findings of *C albicans* may represent true bronchial colonization by *C albicans*, positive sputum culture findings of *A fumigatus* do not represent true bronchial colonization by this fungus. This might be explained because the occurrence of spores in the lung and sputum samples appears to be affected by their size, as suggested by Mullins and Seaton.¹⁶ Thus, *A fumigatus*, with small spores, may be present more frequently in the lung than in samples of respiratory secretions; however, *C albicans* occurs with similar frequencies in the lung and sputum samples.¹⁶

The data currently available on sensitization to *C albicans* are incomplete possibly due to inadequate quality and lack of standardization of fungal

extracts used for skin and serologic tests. Allergic cross-reactivity between products from different fungal species,¹⁷ including *A fumigatus* and *Candida boidinii*,¹⁸ has been described. In our study, the IgE reactivity to *C albicans* was inhibited after serum preadsorption with *C albicans* extract but not *A fumigatus*, demonstrating the specificity of IgE reactivity to *C albicans*. These results are in keeping with those of Roig et al,¹⁰ who did not find allergic cross-reactivity between *A fumigatus* and *C albicans* using immunoblotting.

Our results, unlike those of Wojnarowski et al,⁸ show that sensitization to *A fumigatus* and *C albicans* was not associated with clinical disease. This discrepancy could be due to the fact that the number of lung function tests per patient was higher in the report by Wojnarowski et al.⁸

The IgE immunoresponses against *C albicans* occurred only in *A fumigatus*-sensitized patients and in none of the six atopic patients with negative skin test responses to *A fumigatus*. It has been hypothesized that *A fumigatus* toxins and enzymes injure the airway epithelium in CF patients, permitting leakage of *A fumigatus* allergens into the interstitial spaces.¹⁹ The release of antigenic components of *A fumigatus* may induce a specific IgE production in any CF patient. The cytokines from the Th2-type response induce an inflammatory reaction in the airways, increasing permeability, and this may facilitate access to other fungi colonizing the CF bronchi, such as *C albicans*.

ABPA in patients with CF was first described in 1965.²⁰ ABPA causes complications in 1 to 15% of patients with CF, with up to 23% of the CF patients meeting most but not all of the diagnostic criteria for the disease.⁶ This wide range of prevalence may in part be due to the use of different ABPA diagnostic criteria between centers. In addition, the diagnosis of ABPA in CF patients is often difficult to make because of a number of overlapping clinical and laboratory characteristics.²¹ Different management strategies and the variability in clinical severity of CF patients among hospitals may also explain the different degree of pulmonary colonization by *A fumigatus* and the different risk of developing specific IgE antibodies. The variability in concentrations of exposure to airborne fungal spores in different geographic areas as well as level of suspicion of the disease may also result in a different rate of diagnosis.²² In clinical practice, the diagnosis is often suspected in CF patients who have a positive skin test response and/or serum IgE to *A fumigatus*, together with an elevated total serum IgE, particularly when a pulmonary infiltrate or a decrease in pulmonary function are unresponsive to aggressive antibiotic therapy.

In the present study, we demonstrate IgE to *C albicans* in all 10 ABPA patients and in another 10 patients who had serologic characteristics of ABPA develop, although without clinical suspicion of having the disorder. Nevertheless, we cannot completely rule out that these 10 patients had ABPA because of the nonspecific signs and symptoms of ABPA in this population and the possibility of performing the assessment during asymptomatic intervals. Also, we found a high and significant correlation between IgE to *C albicans* and total serum IgE, the most sensitive marker in ABPA.²³ These data could suggest a contributory role of IgE to *C albicans* in the pathogenesis of ABPA by exacerbating pulmonary infiltrates and inducing eosinophil-mediated inflammatory reaction, as proposed by Roig et al.¹⁰ Thus, IgE to *C albicans*, apart from total IgE and IgE to *A fumigatus*, could be an additional indicator of ABPA activity and could be used as an important aid in the diagnosis and management of this disorder. In our opinion, CF patients with positive IgE to *C albicans* should be screened for ABPA. Since we did not perform skin testing to *C albicans* or serum precipitins to *C albicans*,²⁴ we cannot rule out the possibility of simultaneous occurrence of ABPA and allergic bronchopulmonary candidiasis in some patients, although this has never been documented.

In summary, we found a high prevalence of colonization and sensitization to *A fumigatus* and *C albicans* in CF patients. No correlation was found between the finding of *A fumigatus* in sputum and IgE to *A fumigatus*, whereas only patients who had at least one positive culture finding for *C albicans* during the study had IgE to *C albicans* develop. The sensitization to these fungi is not related to the clinical severity. However, a significant correlation was found between sensitization to both fungi and total serum IgE and total eosinophil in peripheral blood. In addition, IgE to *C albicans* may be an immunologic marker related to the development of ABPA in CF patients.

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