

Incidence of immediate sensitivity to *Aspergillus fumigatus* in a North American asthmatic population

J. L. MALO and R. PAQUIN

Departments of Respiratory Diseases and Haematology, Sacré-Coeur Hospital, Montreal, Canada

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Summary

Allergic bronchopulmonary aspergillosis might be less frequent in North America because the incidence of immediate sensitivity by asthmatics to *A. fumigatus* is less. In order to check this hypothesis, 200 asthmatics were skin tested with two extracts of *A. fumigatus* which had been shown to produce positive reactions in fifty patients who had allergic aspergillosis. Of the asthmatics, 21.5% reacted to the commercial extract by prick testing and 39% by intradermal testing. Using an extract kindly provided by Professor Pepys, 19.5% reacted to a concentration of 1 mg/ml and 31.5% to 10 mg/ml. By the prick method, 21.5% reacted to both extracts. Specific IgE was measured with one of the extracts and a good correlation ($r=0.48$) was found with the size of the prick reaction. The increase in specific IgE was reflected in the increase of total IgE ($r=0.84$). The authors conclude that the incidence of immediate sensitivity to *A. fumigatus* in asthmatic patients in North America is at least equal to that found in the U.K.

Introduction

Allergic bronchopulmonary aspergillosis was first described in the United Kingdom (Hinson, Moon & Plummer, 1952; Pepys *et al.*, 1959; Henderson, 1968) and Europe (Charpin *et al.*, 1967) before there were reports in North America (Patterson & Globert, 1968; Warren & Rose, 1969). The largest series so far published is from England (McCarthy & Pepys, 1971a). Although the exact incidence of this disease among asthmatics in the U.K. and Europe is unknown, there are some doubts that it is as frequent in North America. Several explanations might be given: the quality of the *Aspergillus* extracts used for skin testing and *in vitro* tests, the incidence of immediate and late sensitivity of asthmatics, and the quantity of spores present in the atmosphere.

The present study was conducted in order to determine the incidence of immediate sensitivity to *Aspergillus fumigatus* in a North American asthmatic population.

Correspondence: Dr J. L. Malo, Department of Respiratory Diseases, Sacré-Coeur Hospital, 5400 Gouin West, Montreal, Canada H4J 1C5.

Patients and methods

Two hundred asthmatics seen in a consecutive manner in an asthma clinic were studied. Each of them fulfilled the criteria for asthma as defined by the Ciba Foundation Guest Symposium (1959).

Each of them was skin prick tested (Pepys, 1975) with a routine battery of thirteen commonly inhaled allergens, to wit: alternaria, hormodendrum, house dust, *Dermatophyoides farinae*, mixed tree pollens, grass pollens, ragweed, cat hair, dog hair, horse hair, feathers (Bencard), house dust and *D. farinae* (Hollister-Stier). Skin tests were also done with two different extracts of *A. fumigatus*. These extracts have been shown to produce positive reactions, by the prick testing method, in fifty patients with allergic bronchopulmonary aspergillosis (Malo, Hawkins & Pepys, 1977a). The first extract (Brompton extract) was a lyophilized culture-filtrate preparation provided by Professor J. Pepys and D. J. Longbottom (Department of Clinical Immunology, Cardiothoracic Institute, London S.W.3). This material was prepared in two different concentrations: 1 mg/ml and 10 mg/ml. The second extract was a commercial preparation (Bencard). This extract was used for the prick testing and intradermal methods. The positivity of skin prick and intradermal reactions was assessed according to accepted methods (Pepys, 1975; Chai *et al.*, 1975).

Blood was taken for *in vitro* tests. Total IgE was measured by the PRIST techniques (Ceska & Lundkvist, 1972) using material commercially available from Pharmacia Ltd. Specific IgE values were assessed by the RAST technique as described by Wide, Bennich & Johansson (1967). Discs were coupled with the Brompton extract of *A. fumigatus* and were kindly provided by Professor J. Pepys and Dr J. Longbottom.

Results

Patients

One hundred and seven women and ninety-three men were included in the study. When they were observed, forty-three of them had blood eosinophilia ($\geq 500/\text{mm}^3$) and sixty-three had reversibility of airways obstruction (an increase of 15% or more of peak expiratory flow rate or FEV₁ after three inhalations of an inhaler of salbutamol). Figure 1 shows the histograms of age and age of onset of asthma for these subjects. One hundred and forty nine were atopic (two or more positive prick test reactions with the inhaled allergens used) and fifty-one, non-atopic.

Skin tests

Table 1 gives the results of the skin test reactions with the relevant *A. fumigatus* antigen. A positive reaction was shown by 21.5% of the asthmatics to the commercial antigen by the prick test method and by 39% to the intradermal test. At a concentration of 1 mg/ml, 19.5% had a positive reaction to the Brompton extract and 31.5% at a concentration of 10 mg/ml. By the prick method, 21.5% reacted to both extracts and 31% at least to one or the other of the two.

Specific IgE measurements

Figure 2 shows the mean values (with one standard deviation) of specific IgE for three subgroups of patients: atopic asthmatics with a positive prick test to *A. fumigatus*, atopics with a negative prick test to *A. fumigatus*, and non-atopics. Results for cord blood sera and patients with acute allergic bronchopulmonary aspergillosis are also

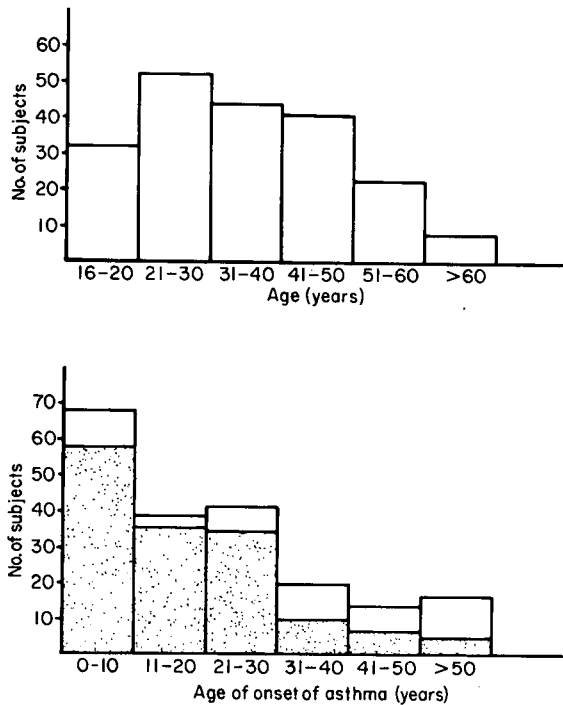


Fig. 1. Histograms of age and age of onset of asthma. The shaded areas represent the number of subjects with two or more positive prick tests to the routine battery of thirteen common inhaled allergens.

Table 1. Results of skin tests

	Prick test			Intradermal test
	Commercial extract	Brompton extract		
		1 mg/ml	10 mg/ml	
1-3 mm	33	32	27	—
4-5	9	5	32	55
> 5 mm	1	2	4	23
Total	43	39	63	78

The results were assessed by measuring the diameter of the weal reaction. In the case of i.d. tests, the result is the diameter of the test made with the *Aspergillus* extract less the diameter of the test with the diluant; a difference of 3 mm and less was considered as non-significant.

given. Atopics with a positive prick test reaction to *A. fumigatus* had significantly ($P < 0.001$) higher values than both atopics with a negative prick test reaction to *A. fumigatus* and non-atopics ($P < 0.001$).

Correlation between skin tests and specific IgE measurements

Table 2 lists the correlations between the prick test results and the specific IgE

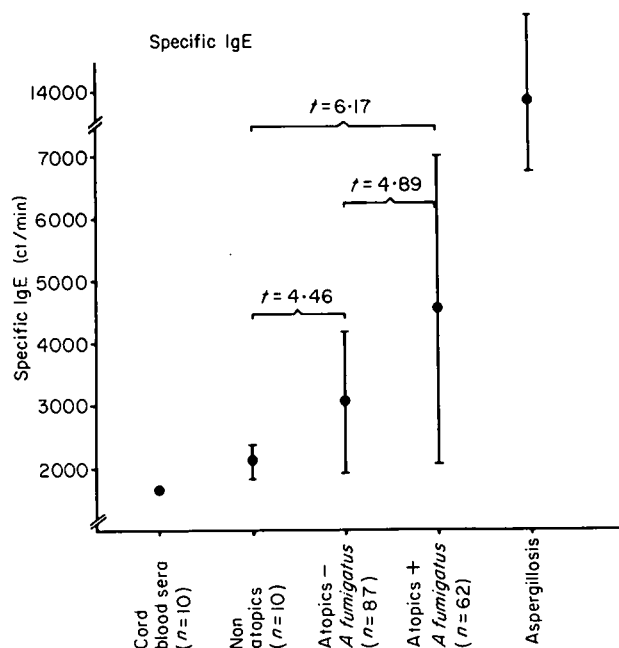


Fig. 2. Mean values with one standard deviation for specific IgE in ct/min for three groups of patients: non atopics, atopics with a negative prick test to *Aspergillus* (Atopics - *A. fumigatus*), and atopics with a positive prick test to *Aspergillus* (Atopics + *A. fumigatus*). Results for cord blood sera and patients with allergic bronchopulmonary aspergillosis (Aspergillosis) are also given. The number (*n*) of subjects in each group is written in brackets. The difference between the mean values of some groups, as assessed by the unpaired *t* test, is shown. All these results are highly significant ($P < 0.001$).

Table 2. Correlation between the results of prick tests and specific IgE levels

Specific IgE score	Prick test score					Correlation
	0.1% - 1% -	0.1% - 1% + + +	0.1% + 1% / + +	0.1% + + 1% + + + + +	0.1% + + + + + 1% + + + + + + +	
0 (< 2500)	71	5	6	0	0	71/82 (87%)
1 (2500-3000)	23	7	2	1	0	10/33 (30%)
2 (3001-4000)	25	4	9	0	0	13/38 (34%)
3 (4001-5000)	5	4	1	7	0	12/17 (71%)
4 (> 5000)	12	5	8	3	2	18/30 (60%)
Correlation	71/136 (52%)	20/25 (80%)	20/26 (77%)	11/11 (100%)	2/2 (100%)	124/200 (62%)

The correlation between the two results is expressed in the following ways: (1) number of subjects with a negative specific IgE score vs number of subjects with a negative prick test; (2) number of subject with a positive specific IgE score vs number of subjects with a positive prick test. Specific IgE levels are given in ct/min. Skin test results were assessed by measuring the weal diameter: +, 1-3 mm; + +, 4-5 mm; + + +, 6-7 mm; + + + +, > 8 mm.

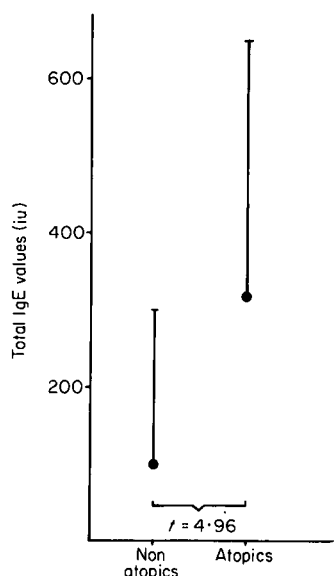


Fig. 3. Values of total IgE in international units (iu) for non atopic and atopic subjects. One standard deviation above the mean is shown. The difference between the mean values, measured by the unpaired t test, is significant at the given t value ($P < 0.001$).

measurements with the same *A. fumigatus* antigen (Brompton extract). Regarding patients who had what is considered as a negative RAST result, most of them (87%) also had a negative skin test. On the other hand, with respect to patients with negative skin reactions, only 52% of them also had a negative RAST result. Nonetheless, most of the subjects with a positive skin test (fifty-three out of sixty-four) also had a positive RAST result. The overall correlation (r) between the size of the prick test and the value of the RAST test was 0.48 ($P < 0.005$).

Total IgE measurements

Values of total IgE were significantly ($P < 0.001$) higher in atopic subjects than in non-atopics. However, as shown in Fig. 3, there was a great overlap of values. A good correlation ($r = 0.84$) was found between the increase in specific IgE and the values of total IgE.

Discussion

The incidence of the immediate sensitivity to *A. fumigatus* varies. In the U.K., Longbottom & Pepys (1964) found that 38% of 238 asthmatics had a positive prick reaction. Hendrick *et al.* (1975) showed that 16% of their 656 asthmatics had a positive prick test. A recent study (Schwartz *et al.*, 1978) compared the incidence of positive prick reactions to mixed *Aspergillus* extracts in asthmatics from the U.K. and the U.S.A. In London 23% reacted and, in Cleveland, 28%. Using a commercial extract of *A. fumigatus* provided by the same company as the one used in the London-Cleveland study, we found a 21.5% incidence of positivity by prick testing and 39% by intradermal testing. It is known that, in the cases of house dust (Spector & Farr, 1974) and *D. farinae* (Stenius *et al.*, 1971), intradermal testing can introduce false positive

reactions which are not confirmed by specific IgE measurements and nasal or bronchial challenges. Although these comparisons have not been examined for *Aspergillus*, there is a possibility that it might also be the case. Using the Brompton extract we found an incidence of positivity of 19.5% with the concentration of 1 mg/ml and of 31.5% with 10 mg/ml. Of our asthmatics, 21.5% reacted to both extracts. Since these two extracts gave positive reactions in all fifty patients with allergic bronchopulmonary aspergillosis (Malo, Hawkins & Pepys, 1977a), we may assume that their quality was good and that a figure of 21.5% would be close to the actual incidence of immediate reactivity in asthmatics.

There was a good overall correlation ($r=0.48$) between the size of the prick test reaction and the values for specific IgE measured with the same extract. This observation agrees with that of Bryant, Burns & Lazarus (1975) who also found, in the case of *A. fumigatus*, that a good correlation ($r=0.51$) between the size of the prick reaction and the RAST values existed.

Although very few patients with a negative RAST test had a positive prick test (11/82), many patients were found to have positive RAST results even if the prick test was negative (65/136). This might have been due to non-specific reactions occurring between the coupled disks and the serum IgE. Indeed, we found that atopic patients with a negative prick test to *A. fumigatus* had significantly higher values for RAST with *A. fumigatus* than non-atopic asthmatics. The general correlation between the positivity of the prick test and the RAST was 62%. This value is lower than the mean of 75% found by Berg & Johansson (1974) in the case of pollens, house dust and animal danders, and than the 76% and 78% agreement, found respectively by Stenius *et al.* (1971), in the case of grass pollens and *D. pteronyssinus*. It is known (Anon., 1977) that depending on the antigen used, the correlation between the prick test and the RAST might be different and this could be the case for *A. fumigatus*.

The values of total IgE were significantly higher in atopic than in non-atopic asthmatics. There was nevertheless a great overlap of values. This confirms the results of Johansson (1967).

Pathogenesis of allergic bronchopulmonary aspergillosis involves both type I and type III reactions. Positive immediate skin reactions and bronchial challenges (McCarthy & Pepys, 1971b), together with increased levels of total IgE (Patterson *et al.*, 1971; Malo *et al.*, 1977b; Imbeau *et al.*, 1978), and specific IgE (Patterson & Roberts, 1974; Turner *et al.*, 1974; Malo *et al.*, 1977b), confirm that an immediate type of reaction contributes to the disease. The presence of late skin and bronchial reactions (McCarthy & Pepys, 1971b), precipitin, and increased specific IgG levels (Patterson & Roberts, 1974; Rosenberg *et al.*, 1978) also show that a type III reaction is involved.

Our study confirms the results of the Cleveland-London survey (Schwartz *et al.*, 1978) that the incidence of immediate sensitivity to *A. fumigatus* of asthmatics is at least as significant in North America as in the U.K. Other factors, contributing to the type III part of the disease (precipitin, specific IgG, levels of *A. fumigatus* in the atmosphere), remain to be investigated.

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