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The evaluation of allergens and allergic diseases in children

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Knowing the incidence of allergic diseases and their relationship with allergens is a crucial requirement for therapeutic judgment. We present our experience on the incidence, clinical features and allergens of the allergic diseases detected by multiple allergosorbent chemiluminescent assay (MAST-CLA) in children from 1997 to 1999 at the Taipei Veterans General Hospital. The incidence of bronchial asthma, allergic rhinitis and atopic dermatitis are significantly different when stratified by age groups. Among the enrolled 2008 patients, 980 (48.8%) patients have positive MAST-CLA results. Of these, 562 (57.3%) are male and 418 (42.7%) are female. A significant increase among patients with positive allergens is also found when stratified by age group. Inhalant allergen is the major allergen detected in our patients. House dust mites *Dermatophagoides pteronyssinus* (Dp) and *Dermatophagoides farinae* (Df), cockroaches, feathers, and dog dander show the highest incidence in the 7- to 12-year-old group. In the fungal group, *Aspergillus* and *Penicillium* also show a significant difference in the incidence among different age groups. Pollen allergens, on a whole, show significant difference in incidence among different age groups. The food allergen group shows variable significant difference in incidence. Crab, milk, and egg white show the highest significant incidence in the 2- to 6-year-old group. These results suggest that the incidence of allergens detected in allergic diseases varies among different age groups.

Key words: Allergens, allergic rhinitis, atopic dermatitis, bronchial asthma

In the last two decades, medical advances have controlled the spread of many infectious diseases. Some diseases have even disappeared. However, increased industrialization has brought severe pollution into the environment, also increasing the incidence of allergic diseases markedly. Therefore, knowing the incidence of allergic diseases and their relationship with allergens is a crucial requirement for therapeutic judgment. Differentiating allergic diseases from nonallergic diseases involves detailed history taking, physical examination and either *in vivo* or *in vitro* testing for relevant allergens. Skin testing is the most widely used diagnostic test for allergies [1]. However, due to its high percentage of false positives and certain contraindication such as skin problems and antihistamine interferences, other diagnostic tests for allergies have been proposed. The multiple allergosorbent chemiluminescent assay (MAST-CLA), radioallergosorbent test (RAST) and Pharmacia CAP test (CAP) all have demonstrated good agreement to skin tests [2-5]. In this paper, we present our experience on the incidence, clinical features and allergens of the allergic diseases detected by MAST-CLA in children at the Taipei Veterans General Hospital from 1997 to 1999.

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Materials and Methods

Patients

From January 1997 to December 1999, 2150 patients with symptoms and signs of bronchial asthma, allergic rhinitis, allergic conjunctivitis, and atopic dermatitis had their blood drawn to check for allergens at the Taipei Veterans General Hospital. Due to incomplete hospital records, 142 patients were excluded from the study. Charts of 2008 patients were reviewed.

MAST-CLA

Blood samples of the patients were sent to the laboratory to have their allergens checked by the MAST-CLA assay (MAST-Immunosystems, Mountain view, California, USA) test. MAST-CLA can simultaneously measure 35 specific IgE antibodies. The allergens chosen for the Taipei Veterans General Hospital were common inhalant allergens, mold, pollen and food allergens. MAST-CLA requires 1.3 mL of serum and is completed within 24 h. The system consists of a long pipette with 38 parallel

cellulose threads strung ladder-fashion from top to bottom. Each rung has a different allergenic extract covalently bound to it. Positive and negative controls are also included in the system [6].

Sample analysis

Each of the patient's serum is drawn up into the pipette and incubated overnight. After washing, the pipette is filled with a solution of anti-IgE, labeled with a luminescent substance and incubated for another 4 h. After washing and addition of a photoreagent mixture, a band of light is emitted from threads where IgE has bound. The pipettes are then exposed onto a Polaroid film, and the degree of light emitted is measured by densitometry and expressed in volts. The quantity of IgE present is proportional to this voltage minus the voltage of the negative control. The result of each thread is then expressed in four classes: 0 = nondetectable, 1 = low, 2 = moderate, 3 = high, 4 = very high. Positive results are defined as upper than class 2.

Statistical analysis

Statistical significance is analyzed by the Student's t-test, Fisher's exact test and Chi-square analysis. A p value of less than 0.05 is considered as statistically significant.

Results

For a period of 2 years, 2008 patients with symptoms and signs of bronchial asthma, allergic rhinitis and atopic dermatitis had their blood drawn to check for allergens at the Taipei Veterans General Hospital. Their ages ranged from 7 months to 18 years, with a mean of 6.7 ±3.96 years. Patients were grouped into seven disease subgroups: bronchial asthma (BA), allergic rhinitis (AR), atopic dermatitis (AD), bronchial asthma + allergic rhinitis (BA+AR), bronchial asthma + atopic dermatitis (BA+AD), allergic rhinitis + atopic dermatitis (AR+AD), and bronchial asthma + allergic rhinitis + atopic dermatitis (BA+AR+AD). Table 1 shows the cumulative incidence of allergic disease among different age groups. The incidence of BA, AR, AD, BA+AR, BA+AD, AR+AD and BA+AR+AD show significant difference when compared among different age groups. Bronchial asthma shows the highest incidence in the 2- to 6-year-old group, while allergic rhinitis and atopic dermatitis show the highest incidence in the 7- to 12-year-old group.

Table 2 compares the incidence of positive allergens by MAST-CLA among different age groups. Among the enrolled 2008 patients, 980 (48.8%) patients had positive MAST-CLA results. Of these, 562 (57.3%) were male and 418 (42.7%) were female. When the incidence of positive MAST-CLA was stratified by age group, a statistically significant increase proportional to age was found.

The incidence of clinical symptoms and signs found in allergic patients is shown in Table 3. Sneezing, with the highest incidence of 66.1%, was the most common symptom reported by the patients. It is followed by nose

Table 1. Cumulative incidence of allergic diseases among different age groups

Subgroup	<2 y/o	2-6 y/o	7-12 y/o	13-18 y/o	p^a
BA (n = 373)	11	216	125	21	<0.001
AR (n = 404)	9	170	191	34	<0.001
AD (n = 125)	17	28	48	32	<0.005
BA+AR (n = 754)	2	298	383	71	<0.001
BA+AD (n = 69)	19	35	12	3	<0.001
AR+AD (n = 86)	26	49	7	4	<0.001
BA+AR+AD (n = 197)	4	136	46	11	<0.001
Total (n = 2008)	88	932	812	176	

Abbreviations: BA = bronchial asthma, AR = allergic rhinitis, AD = atopic dermatitis
[a] p value compares the incidence of each allergic disease among different age groups.

Table 2. Incidence of positive allergens by MAST-CLA among different age groups

	<2 y/o	2-6 y/o	7-12 y/o	13-18 y/o	p^a
Total no. of patients (n = 2008)	88	932	812	176	
No. of positive MAST-CLA (n = 980)	24	356	476	124	
% of positive MAST-CLA	27.3	38.2	58.6	70.5	<0.001

Abbreviations: MAST-CLA = multiple allergosorbent chemiluminescent assay
[a] p value compares the percentage of positive MAST-CLA among different age group.

Table 3. Clinical symptoms and signs found in 2008 patients with allergic diseases

Symptom/sign	No. of patients (%)
Sneezing	1327 (66.1)
Nose itching	1312 (65.3)
Nasal obstruction	1192 (59.4)
Cough	1028 (51.2)
Rhinorrhea	867 (43.1)
Shortness of breath	790 (39.3)
Throat injection	580 (28.9)
Eye itching	398 (19.8)
Eczema	345 (17.2)

itching, nasal obstruction, cough, rhinorrhea, shortness of breath, throat injection, eye itching and eczema.

Table 4 compares different allergens detected among the age groups. Allergens were classified into four groups: inhalant allergens, mold, pollen and food allergen. The inhalant allergen was the major allergen detected in our patients. In the inhalant allergen group, house dust mites *Dermatophagoides pteronyssinus* (Dp) and *D. farinae* (Df), cockroaches, feathers, and dog dander show the highest significant incidence in the 7- to 12-year-old group. In the mold group, *Aspergillus* and *Penicillium* also show a significant difference in incidence among different age groups. Pollen as a whole showed a significant difference in incidence among different age groups. However, there is no significant difference in incidence if allergens are compared separately. The food allergen group shows a significant difference in incidence among vegetable mix, crab, shellfish mix, egg white, milk, shrimp, brewer's yeast, and peanut. Crab, milk and egg white show the highest incidence in the 2- to 6-year-old group.

Table 5 compares the incidence of allergens detected among allergic diseases. The inhalant allergen is the most common allergen detected in the BA, AR, BA+AR, AR+AD and BA+AR+AD groups. The food allergen is the second most common allergen detected. All the data is statistically significant when allergens are compared among themselves.

Discussion

Allergic diseases have increased markedly both in incidence and severity in the last two decades in both industrialized countries and in developing countries such as Taiwan [7,8]. Since the first proposal by Von Pirquet in 1902, allergic diseases have gradually become an important issue of public health [9]. It was not until 1922 when Coca *et al* suggested that patients with allergic diseases, such as bronchial asthma, allergic rhinitis, and atopic dermatitis, not only have a hereditary tendency, but also are hypersensitive to environmental allergens [10]. To investigate a patient with a suspected allergic disease, a detailed clinical history, thorough physical examination and confirmatory diagnostic testing are required [2-5]. In this study, we used MAST-CLA to detect allergens of our patients.

Among the enrolled 2008 patients, 980 (48.8%) patients have positive MAST-CLA results. This is a relatively low positive rate when compared with others [11,12]. We believe the low positive rate is related to our patients' age and our food allergens. Our patients have a lower mean age of 6.7 ±3.96 years. If we consider only those patients older than 6 years of age, then the positive MAST-CLA rate would be 60.7%, which is much higher than 48.8%. Secondly, Lo *et al* has previously pointed out that the positive rate of food allergens is much lower than that of aeroallergens (35.3% vs 82.8%) [12]. The different results might be related to the quality of the extract and the difference of correspondent allergens employed in the tests.

Our results show that the predilection for allergic diseases varies statistically among different age groups. The incidence of bronchial asthma in patients aged or younger than 6 years old is 60.8%. This result is similar to that of Wilken-Jensen, lower than that reported by Ryssing or Lue [13-15] and higher than that in our previous reports [16,17]. The incidence of allergic rhinitis is 47.2% in the 7- to 12-year-old group. This is similar to the results reported by Freeman [18]. The incidence of atopic dermatitis is the highest in the 7- to 12-year-old group. These results are not compatible with many previously published reports, including that proposed by Hill *et al*, who pointed out that the majority of atopic dermatitis occurs in infancy [19]. This discrepancy might be secondary to a few MAST tests performed in infancy with clinically significant atopic dermatitis in this study.

When the incidence of positive allergens is compared in different age groups, a statistically significant increase in incidence is found. This increase in positive allergens is related to the immature immunity of younger patients [20]. In this retrospective study, no skin test was done. Data about sensitivity, specificity or efficiency was unavailable. Therefore, we cannot make any conclusion on the recommended age for performing MAST-CLA.

The house dust mites, Dp and Df, have been identified as the main sources of house dust allergens [21,22]. These allergens are considered to be the most important causative allergen in perennial rhinitis and extrinsic asthma all over the world, including in asthmatic children in Taiwan. [23,24]. Our study shows

Table 4. Comparison of different allergens detected among different age groups

Allergen	<2 y/o	2-6 y/o	7-12 y/o	13-18 y/o	p^a
Inhalant allergen (n = 2596)	56	589	1592	359	<0.001
House dust (n = 600)	8	192	344	56	<0.001
Mite (Df) (n = 936)	24	324	476	112	<0.001
Mite (Dp) (n = 950)	24	338	472	116	<0.001
Cockroaches (n = 309)	0	49	212	48	<0.001
Feathers (n = 58)	0	4	42	12	<0.001
Dog dander (n = 62)	0	18	37	7	<0.001
Cat dander (n = 24)	0	7	9	8	NS
Fungus (n = 84)	0	0	61	23	<0.001
Candida albicans (n = 20)	0	0	11	9	NS
Aspergillus (n = 21)	0	0	16	5	<0.05
Cladosporium (n = 22)	0	5	12	5	NS
Penicillium (n = 17)	0	0	13	4	<0.05
Alternaria (n = 9)	0	0	9	0	NS
Pollen (n = 58)	0	0	18	40	<0.005
Ragweed (n = 5)	0	0	0	5	NS
Grass mix (n = 10)	0	0	4	6	NS
Pine mix (n = 4)	0	0	0	4	NS
Cotton (n = 4)	0	0	0	4	NS
Eucalyptus (n = 3)	0	0	0	3	NS
Mulberry mix (n = 13)	0	0	5	8	NS
Bermuda grass (n = 13)	0	0	6	7	NS
Pigweed mix (n = 6)	0	0	3	3	NS
Food allergen (n = 642)	16	178	334	114	<0.001
Corn (n = 4)	0	0	0	4	NS
Wheat (n = 17)	0	6	5	6	NS
Vegetable mix (n = 21)	0	0	17	4	<0.05
Crab (n = 141)	0	62	61	18	<0.001
Shellfish mix (n = 74)	0	29	37	8	<0.001
Codfish (n = 13)	0	5	8	0	NS
Pork (n = 14)	0	5	4	5	NS
Beef (n = 5)	0	0	0	5	NS
Egg white (n = 66)	4	33	16	13	<0.001
Egg yolk (n = 22)	0	10	0	12	NS
Milk (n = 79)	4	45	21	9	<0.001
Shrimp (n = 130)	0	53	65	12	<0.001
Brewer's yeast (n = 25)	0	6	15	4	<0.05
Soybean (n = 28)	0	11	8	9	NS
Peanut (n = 108)	0	26	77	5	<0.001

Abbreviations: D = *Dermatophagoides farinae*; Dp = *Dermatophagoides pteronyssinus*; NS = non-significant
[a] *p* value compares the incidence of allergens detected among different age groups.

similar results, with Dp being the most prevalent allergen, and with inhalant allergens being the most common allergen in bronchial asthma and allergic rhinitis.

Allergy to fungal spores is an important cause of disease in many atopic patients. In the present study, fungal allergens are found mainly in children older than 7 years of age, with *Cladosporium* being the most commonly found fungal allergen. As a whole, fungal allergens are found most commonly in patients with atopic dermatitis.

Different in geographic locations and seasons, allergenic pollens are discharged from wind-pollinated plants. Even though wind-pollinated plants are much fewer than insect-pollinated plants, they discharge large amounts of light-weighted pollens which are dispersed by wind currents. We identified pollen mainly in children older than 7 years of age, with Bermuda grass and Mulberry being the most common pollen.

Food allergens can induce IgE antibodies that are considered to be responsible for atopic reactions [25, 26]. The present study finds shrimp to be the most

Table 5. Relationship between allergens and allergic diseases: overall results

Subgroup	Inhalant allergen (n = 2596)	Fungus (n = 84)	Pollen (n = 58)	Food allergen (n = 642)	p^a
BA (n = 689)	607	15	10	57	<0.001
AR (n = 619)	521	16	12	70	<0.001
AD (n = 249)	35	5	4	205	<0.001
BA+AR (n = 1095)	976	31	21	67	<0.001
BA+AD (n = 198)	88	3	2	105	<0.001
AR+AD (n = 190)	109	4	3	74	<0.001
BA+AR+AD (n = 340)	260	10	6	64	<0.001

Abbreviations: BA = bronchial asthma; AR = allergic rhinitis; AD = atopic dermatitis
[a] *p* value compares the incidence of allergens among allergic diseases.

common food allergen, followed by peanut and egg white. Food allergens in our study are mostly associated with atopic dermatitis. Bronchial asthma is associated with 57 food allergens. Adler *et al* reported the inclusion of food specific IgE antibodies together with inhalant antibodies in MAST-CLA for use in atopic asthmatics might be misleading [27]. Therefore, the relationship between food allergens and bronchial asthma should be questioned.

In the last two decades, medical advances have controlled the spread of many infectious diseases. Some diseases have even disappeared. However, increased industrialization has brought severe pollution into the environment, also increasing the incidence of allergic diseases markedly [28]. Therefore, knowing the incidence of allergic diseases and their relationship with allergens are necessary in managing these diseases. The result of this study may be helpful in educating patients about allergen avoidance and environmental care.

Acknowledgments

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