Effect of Age on Allergen Responses of Allergic Patients in Southern Taiwan

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To survey airborne and food allergen patterns in southern Taiwan and to analyze the effect of age on response to different allergens, we tested samples from 4,411 allergic patients at Kaohsiung Medical University Hospital using the MAST-CLA test (new Taiwan panel). A total of 2,212 (50.1%) samples showed a positive response. We grouped allergic patients into five age groups. Milk and egg white were the main food allergens in the younger groups (< 3 years old and 3–6 years old). Shrimp, crab, and shellfish were the main allergens in the groups aged 7–12, 13–18, and more than 18 years. Among airborne allergens, house dust and mites *Dermatophagoides farinae* and *D. pteronyssinus* were the main allergens in all age groups, whereas the frequency of response to cockroach allergen was low in the group aged less than 3 years, but increased in the other age groups. There was a sharp increase in the frequency of response to airborne allergens after 3 years old and a sharp decrease in response to food allergens. Among subjects allergic to both airborne and food allergens, there was a positive MAST-CLA rate of 19.9% to 26% (all five age groups, no significant difference). When we compared our results with those from Taipei Veterans General Hospital in northern Taiwan, there were significant differences for yeast, peanut, feather mix, dog dander, cockroach, *D. farinae* and *D. pteronyssinus* allergens ($p < 0.01$). These differences were probably caused by differences in patient location, patient age, disease patterns and allergen panels.

Key Words: MAST-CLA test, allergic patterns, southern Taiwan, food allergens, airborne allergens
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Atopic diseases are increasingly common in developing and developed countries, and the medical cost has become a large burden for families and for society in general [1]. If atopic subjects can be detected in early childhood, it might be possible to stop or slow the atopic march [2]. Increased industrialization has brought severe pollution that has increased the incidence of allergic disease markedly.

The detection of allergen-specific immunoglobulin E (IgE) antibodies in serum is easy and non-invasive and helps doctors to diagnose atopic subjects [3]. The MAST-CLA (multiple allergosorbent chemiluminescent assay) test, radioallergosorbent test, and Pharmacia CAP test all have good agreement with the traditional skin test [4–7]. The MAST-CLA test is an *in vitro* assay to simultaneously screen for specific IgE antibodies to 35 allergens (15 food allergens and 20 airborne allergens) in the serum of subjects suspected to be atopic. The MAST-CLA panel has been used routinely in immunology laboratories in Taiwan for 10 years. Since February 2001, a new MAST-CLA Taiwan panel, with *Dermatophagoides farinae* and *D. pteronyssinus* allergens at the bottom of the test pipette and the addition of citrus allergen, has replaced the old one.

In Taiwan, data from the MAST-CLA test for several allergens in allergic children have been reported in northern Taiwan [4,7–11], but very few data for southern Taiwan have been documented [12]. To our knowledge, no paper has been published since the implementation of the new MAST-CLA panel in Taiwan. The purpose of this study was

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to investigate allergen frequency in southern Taiwan, to determine the effect of age on allergen responses using the new MAST-CLA panel in allergic patients at the Kaohsiung Medical University Hospital (KMUH), and to compare the incidence of food and airborne allergens with that reported from Taipei Veterans General Hospital (VGH) in northern Taiwan.

Materials and Methods

Between February 2001 and April 2002, we took serum samples from 4,411 enrolled patients (mean age, 17.1 ± 11.1 years; range, 0–92 years) with signs and symptoms of bronchial asthma, allergic rhinitis, allergic conjunctivitis, or atopic dermatitis at KMUH. The MAST-CLA (Hitachi Chemical Diagnostics Inc, Mountain View, CA, USA) new Taiwan Allergy Panel, including common inhalant allergens, mold, pollen, and food allergens, was used to assess the frequency of responses to the allergens. MAST-CLA requires 1.3 mL of serum and is completed within 24 hours. The system consists of a long pipette with 38 parallel cellulose threads strung ladder-fashion from top to bottom. Each thread has a different allergenic extract covalently bound to it. Positive and negative controls are also included in every test pipette.

Each patient's serum was drawn up into the pipette and incubated overnight. After washing, the pipette was filled with a solution of enzyme-labeled goat anti-human IgE and incubated for another 4 hours. After washing and the addition of a photoreagent mixture, a band of light is emitted from threads where IgE has bound. After 10 minutes' incubation, the pipette was placed in a luminometer to measure the amount of light emitted by each thread. The quantity of IgE present is proportional to the number of luminescence units (LUs) of each allergen minus the LUs for the negative control. The result from each thread is then expressed as one of six classes: 0 = undetectable (net LUs, 0–11), ± = very low (net LUs, 12–26), 1 = low (net LUs, 27–65), 2 = moderate (net LUs, 66–142), 3 = high (net LUs, 143–242), and 4 = very high (net LUs, > 242). Positive results were defined as greater than class 2. We grouped positive samples by age into five groups: less than 3 years, 3–6 years, 7–12 years, 13–18 years, and more than 18 years.

Statistical analysis

Results were analyzed for statistical significance using the Chi-squared test. A p value of less than 0.05 was considered statistically significant.

Results

Frequency of allergens in different age groups

Among the enrolled patients, 2,212 (50.1%) had positive MAST-CLA results. Of these, 1,263 (57.1%) were male and 949 (42.9%) were female.

The frequencies of response to 16 food allergens are shown in Table 1. For the group aged less than 3 years, the most common allergen was milk (65.3%) followed by egg white (33.3%). This decreased to 5% and 2.8%, respectively, for the 7–12-year-old group. In this group, crab, shellfish mix, and shrimp were the three most common food allergens (12.8%, 10.1%, and 13.4%, respectively). Among seafood allergens, shrimp was the most common in all five age groups, followed by crab. The newly added citrus allergen showed low frequency in all age groups.

Positive MAST-CLA responses to 20 airborne allergens are shown in Table 2. Among all age groups, the most common airborne allergens were the mites *D. farinae* and *D. pteronyssinus*, followed by house dust and cockroach mix. The frequencies of *D. farinae* allergen increased from 42.7% in the group aged less than 3 years to 95.5% in the 7–12-year-old group and remained high in the 13–18-year-old group (89.9%) and the group aged more than 18 years (84.1%). The frequencies of *D. pteronyssinus* allergen increased from 46.7% in the group aged less than 3 years to 92.1% in the 7–12-year-old group and remained high in the 13–18-year-old group (89.9%) and the group aged more than 18 years (75.6%).

For further analysis of the effect of different patterns of food and airborne allergens, we divided patients into three groups: allergic to food allergen only, allergic to airborne allergen only, and allergic to both food and airborne allergens. Table 3 gives the frequencies of these allergies in the five age groups. The frequencies of allergy to airborne allergens increased rapidly after the age of 3 years and were significantly different among the five age groups ($p < 0.001$). The frequencies of allergy to food decreased rapidly after 3 years old and were significantly different among the five age groups ($p < 0.001$). The frequencies of allergies to both airborne and food allergens remained steady at 19.9% to 26% for all five age groups ($p > 0.05$).

Comparison with VGH: food allergens

Comparison of food allergen frequencies between our age-matched (< 18 years) samples and the results in Lee et al's report from northern Taiwan are shown in Table 4 [8]. Positive frequencies of food allergens between these two studies showed significant differences for peanut and brewer's yeast ($p < 0.01$), and for crab, shellfish mix, wheat,

Table 1. Frequencies (%) of response to food allergens among five age groups

	< 3 yr	3–6 yr	7–12 yr	13–18 yr	> 18 yr	*p**
Total, *n*	76	308	776	310	742	
Citrus mix	0.0	1.6	0.5	0.7	1.9	NS
Corn	0.0	1.0	0.8	0.3	1.6	NS
Wheat	1.3	1.3	0.4	0.3	2.0	< 0.05
Vegetable mix	0.0	2.9	2.5	1.3	1.9	NS
Crab	2.6	10.8	12.8	9.4	6.5	< 0.001
Shellfish mix	1.3	9.8	10.1	7.8	5.8	< 0.001
Shrimp	5.3	11.1	13.4	14.3	13.9	NS
Codfish	2.6	4.2	2.5	2.0	2.7	NS
Pork	2.6	0.7	0.4	0.0	0.7	NS
Beef	0.0	0.0	0.0	0.0	0.4	NS
Milk	65.3	19.6	5.0	1.3	1.6	< 0.001
Yeast, brewer's	1.3	0.3	1.0	0.3	0.4	NS
Soybean	2.6	3.3	1.6	0.7	3.8	< 0.05
Peanut	4.0	4.2	1.2	0.3	2.4	< 0.01
Egg yolk	4.0	1.0	0.8	0.0	0.5	< 0.01
Egg white	33.3	12.7	2.8	0.7	0.9	< 0.001

*Comparison of the percentage of positive MAST-CLA results among different age groups. NS = non-significant.

Table 2. Frequencies (%) of response to airborne allergens among five age groups

	< 3 yr	3–6 yr	7–12 yr	13–18 yr	> 18 yr	*p**
Total, *n*	76	308	776	310	742	
Pine mix	1.3	1.6	0.9	0.7	1.9	NS
Cotton/willow	1.3	0.3	0.4	0.3	1.8	NS
Eucalyptus	0.0	0.3	0.5	0.0	1.4	NS
Mulberry mix	0.0	0.3	0.8	0.3	1.8	NS
Grass mix	0.0	2.9	2.7	2.0	7.0	< 0.001
Bermuda grass	0.0	3.6	3.4	2.0	7.3	< 0.001
Ragweed mix	0.0	1.6	1.7	0.7	3.6	< 0.01
Pigweed mix	0.0	0.7	0.6	0.3	2.6	< 0.01
Alternaria	0.0	0.7	1.9	2.3	1.5	NS
Aspergillus	1.3	0.3	1.8	2.6	2.3	NS
Candida	0.0	0.0	1.9	3.9	3.8	< 0.01
Cladosporium	0.0	0.0	1.2	1.6	1.6	NS
Penicillium	2.6	0.3	1.7	4.9	4.7	< 0.001
Feather mix	0.0	1.6	1.0	1.0	1.5	NS
Cat dander	2.6	2.6	4.1	6.2	4.3	NS
Dog dander	5.3	6.5	3.6	14.6	9.6	< 0.001
Cockroach mix	2.6	13.1	22.4	25.0	23.7	< 0.001
House dust	25.3	58.5	60.6	63.0	32.9	< 0.001
D. pteronyssinus	46.7	82.0	92.1	89.9	75.6	< 0.001
D. farinae	42.7	88.2	95.5	89.9	84.1	< 0.001

*Comparison of the percentage of positive MAST-CLA results among different age groups. NS = non-significant.

codfish, egg yolk, pork, and beef ($p < 0.05$).

Comparison with VGH: airborne allergens

The frequencies of airborne allergens in our age-matched (< 18 years) samples are compared with those in Lee et al's report from northern Taiwan in Table 5 [8]. There were significant differences in the frequencies of airborne allergens for *D. farinae* and *D. pteronyssinus*, cockroach mix, feather mix, and dog dander ($p < 0.01$), and cat dander, Bermuda grass, and *Cladosporium* ($p < 0.05$).

Table 3. Frequencies (%) of allergies to airborne allergen(s), food allergen(s), and both allergens among the five age groups

	< 3 yr	3–6 yr	7–12 yr	13–18 yr	> 18 yr	*p**
Total, *n*	76	308	776	310	742	
Airborne only	33.0	69.5	75.5	73.0	77.4	< 0.001
Food only	41.4	5.4	1.8	1.0	2.7	< 0.001
Airborne and food	25.6	25.1	22.7	26.0	19.9	NS

*Comparison of the percentage of positive MAST-CLA results among different age groups. NS = non-significant.

Table 4. Comparison of frequencies (%) of response to food allergens in patients less than 18 years old, between Taipei Veterans General Hospital (VGH) and Kaohsiung Medical University Hospital (KMUH)

	VGH	KMUH	*p**
Total	980	1,470	
Corn	0.4	0.7	NS
Wheat	1.7	0.6	< 0.05
Vegetable mix	2.1	2.3	NS
Crab	14.4	11.0	< 0.05
Shellfish mix	7.6	10.5	< 0.05
Shrimp	13.3	12.7	NS
Codfish	1.3	2.7	< 0.05
Pork	1.4	0.5	< 0.05
Beef	0.5	0.0	< 0.05
Milk	8.1	10.3	NS
Yeast, brewer's	2.6	0.8	< 0.01
Soybean	2.9	1.8	NS
Peanut	11.0	1.8	< 0.01
Egg yolk	2.2	1.0	< 0.05
Egg white	6.7	5.8	NS

*Comparison of the percentage of positive MAST-CLA results between the two hospitals. NS = non-significant.

Table 5. Comparison of frequencies (%) of response to airborne allergens in patients less than 18 years old, between Taipei Veterans General Hospital (VGH) and Kaohsiung Medical University Hospital (KMUH)

	VGH	KMUH	*p**
Total	980	1,470	
Pine mix	0.4	1.0	NS
Cotton/willow	0.4	0.4	NS
Eucalyptus	0.3	0.3	NS
Mulberry mix	1.3	0.5	NS
Grass mix	1.0	1.6	NS
Bermuda grass	1.3	2.9	< 0.05
Ragweed mix	0.5	1.4	NS
Pigweed mix	0.6	0.5	NS
Alternaria	0.9	1.6	NS
Aspergillus	2.1	1.6	NS
Candida	2.0	1.8	NS
Cladosporium	2.2	1.0	< 0.05
Penicillium	1.7	2.1	NS
Feather mix	5.9	1.9	< 0.01
Cat dander	2.4	4.2	< 0.05
Dog dander	6.3	10.1	< 0.01
Cockroach mix	31.5	19.7	< 0.01
House dust	61.2	59.1	NS
D. pteronyssinus	95.5	89.7	< 0.01
D. farinae	96.9	86.7	< 0.01

*Comparison of the percentage of positive MAST-CLA results between the two hospitals. NS = non-significant.

Discussion

In developed countries, atopic diseases are very common: about 30% to 40% of the general population is atopic [1]. In the USA, the prevalence and severity of asthma has increased in the last 20 years, and the greatest increase has been seen among children and young adults living in inner cities. The reasons for this increase are obviously complex, but include environmental exposure to allergens and pollutants, changing patterns of medication, and the psychosocial stresses of living in poor inner-city neighborhoods [13].

An easy *in vitro* test is necessary to help physicians screen for atopic individuals. The MAST-CLA test is a semi-quantitative assay that can screen for specific IgE antibodies to 36 common allergens. *D. farinae*, *D. pteronyssinus*, and house dust were the three most common allergens in our study. Although the allergen frequencies in this study and the effect of age on allergic responses are similar to data from northern Taiwan for most allergens [8], there were significant differences for brewer's yeast, peanuts, feather mix, dog dander, cockroach mix, *D. farinae* and *D. pteronyssinus* ($p < 0.01$). These differences were probably caused by differences in population and age group between VGH (6.7 ± 4.0 years) and KMUH (17.1 ± 11.1 years), differences in living environment and food between northern and

southern Taiwan, disease patterns, and the re-arrangement of allergens in the new Taiwan Allergy Panel. Hill and Sulzberger reported that most atopic dermatitis occurs in infancy [14]. The effects of re-arrangement of allergens may be the major cause of the differences in peanut (the last allergen in the old panel, which might have led to problems from incomplete washing) and feather mix (next to the cockroach mix in the old panel, which might have led to contamination by carry-over to produce a false-positive result). Our results indicate that the highest frequency of specific IgE antibody to milk was in younger children (< 3-year-old group), and the frequencies gradually decreased as age increased. These results are similar to those of Tsai and Chen [15]. This phenomenon is correlated with the "grow out" theory proposed by Sampson [16].

Several previous reports suggest that the appearance of specific IgE antibody to egg white and cow's milk in early life and the persistent appearance of specific IgE antibody to cow's milk might be a predictor of the atopic march, and avoidance of these food allergens in infancy could stop or slow atopic progression [17–21]. Dynowska et al [22] and Kotaniemi-Syrjanen et al [23] observed that the appearance of specific IgE antibodies to airborne allergens in infancy provided evidence of atopy. The appearance of specific IgE antibodies to *D. farinae* and *D. pteronyssinus*, house dust, and cockroach among older children and adults is correlated with many atopic diseases such as atopic dermatitis, asthma, and allergic rhinitis [8–10,24].

In Lee et al's study of fungal allergens, *Cladosporium* was the most commonly found fungal allergen [8], although in our study, it was the least common fungal allergen. Our study showed that the frequencies of allergy to airborne allergen(s) increased as patient age increased. However, the frequencies of allergy to food allergen(s) decreased as patient age increased. This trend was probably caused by changes in living habits. The constant frequencies of allergy to both airborne and food allergens in all five age groups suggested that this kind of allergy (particularly in infancy) should be kept under regular surveillance to prevent further development.

Most atopic individuals are unaware of their disease, even when they suffer anaphylactic shock [25]. Tang et al reported that the results of total IgE were not reliable in diagnosis of allergy [7]. We suggest that babies at high risk (i.e. those who come from atopic families) should be screened for specific IgE in infancy, then periodically rechecked (every 3 or 4 months in infancy, every 6 months in older children) using the MAST-CLA test. Doctors would then be able to identify suspected atopic babies and the causative allergens. This would allow the use of proper treatments and home education for parents. Finally, the atopic march might be stopped or slowed and the atopic population reduced.

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南台灣過敏患者之年齡對過敏原反應的影響

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過敏病患在各個國家均為普遍發生之疾病 (發生率為 **30%** 至 **40%**)，如何檢視出此類病患並給予適當治療與衛教實為困難煩索之工作。幸而體外檢測出的特異性免疫球蛋白 **E** 可提供相當可靠的診斷依據給醫師。**MAST-CLA** 即為其中一種普遍使用之體外檢驗，本部免疫室於 **MAST-CLA** 新版試劑上市後，收集高雄醫學大附設中和紀念醫院有過敏症狀之患者之受測檢體 **4,411** 件 (年齡自 **0** 歲至 **92** 歲)，其中 **MAST-CLA** 呈陽性反應 ($\geq$ **2**) 者 **2,212** 件 (**50.1%**，其中男性 **57.1%**，女性 **42.9%**)，若依患者年齡分成五組後，統計各種過敏原之分布頻率，結果顯示小於 **3** 歲之幼兒主要過敏原為牛奶及蛋的蛋白，待年齡增長後過敏原逐漸以吸入性過敏原 (美洲塵蟎，歐洲塵蟎，蟑螂) 為主，食入性過敏原也轉變成海產類為主。與北台灣李等報告之結果相比較時，本研究中酵母菌，花生，羽毛，蟑螂，美洲塵蟎，歐洲塵蟎等過敏原之分布頻率有顯著之偏低 ($p < 0.01$)，狗毛過敏原卻顯著偏高 ($p < 0.01$)。此等差異可能是因不同的族群，患者的年齡，疾病的種類或不同的過敏原組合造成的結果。在 **5** 個不同年齡組中，同時對吸入性及食入性過敏原，產生特異性免疫球蛋白 **E** 的患者比率，均維持在 **19.9%** 至 **26%** 之間，這些患者若能早期診斷出來，並施以適當的治療與照顧，或許可以減緩其它過敏症狀的發生。定期的複檢亦可監控其致病過敏原種類的改變。

關鍵詞：多項過敏原篩檢法，南台灣，食入性過敏原，吸入性過敏原
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