

Allergen Sensitization to Aeroallergens Including *Blomia tropicalis* among Adult and Childhood Asthmatics in Thailand

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Sensitization to allergens, particularly to house dust mites and cockroaches is the major risk factor for the development of asthma.¹⁻³ Degree of sensitization to aeroallergens correlates with the prevalence of asthma⁴ and severity⁵ as well as bronchial reactivity,⁶ particularly in childhood asthma. Moreover, the allergen avoidance leads to a marked reduction of symptoms, to an improvement of nonspecific bronchial reactivity⁷⁻¹⁰ with ability to reduced requirement of inhaled corticosteroids.¹¹ Recent investigation in Thailand indicated that allergen sensitization is the most important risk factor for childhood asthma.¹² The incidence of allergen sensitization among childhood asthmatics in Thailand (67%) is similar to the rates reported around the world.¹³ However, the data on allergen sensitization in adult asthmatics in Thailand are limited.

Recent study demonstrates that *Blomia tropicalis* (*Bt*), a storage

SUMMARY To study prevalence of allergen sensitization among asthmatics in Thailand, skin prick tests (SPT) were performed in 84 pediatric, 71 adult asthmatics and 71 adult volunteers. Allergen extracts used for testing included common allergens in Thailand and in Singapore. The incidence of positive SPT to any allergen among the three groups (childhood, adult patients and adult controls) were 64.3%, 43.7% and 35.2%, respectively. *Dermatophagoides* were the most common allergens sensitized by both pediatric (58.3%) and adult asthmatics (40.8%). Twenty-four children (28.6%) and 8 adult patients (11.3%) were sensitized to storage mites (*Blomia tropicalis* and/or *Austrogllyphagus malaysiensis*). All patients sensitized to *Blomia tropicalis* were sensitized to *Dermatophagoides*. Twenty-seven percent and 15.5% of childhood and adult asthmatics were sensitized to cockroach allergens. The rates of sensitization to oil palm pollen in childhood and adult asthmatics were 8.3% and 5.6%, respectively. Sensitization to other pollens and spores were less than 5%. This study confirms the importance of *Dermatophagoides* among Thai asthmatics.

mite is an important mite species causing sensitization in asthmatic patients in South America,¹⁴ and in southern parts of USA.^{15,16} In Asia, *Bt* is the major mites causing sensitization among atopic patients in Singapore,¹⁷ Malaysia¹⁸ and Indonesia.¹⁹ Our experience in mite identification in Thailand indicated that *Bt* was rarely found in dust collected from homes in Thailand.²⁰ In addition, Resam fern spore (RF), unusual mold spores and pollen from oil palm tree, an important economic

plant for this region, have been reported to be important aeroallergens in Southeast Asia.²¹

The objectives of this study are (1) to study the prevalence of sensitization to common aeroallergens among childhood and adult asthmatics in Thailand, and (2) to

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study the incidence of sensitization to *Bt* and newer aeroallergens, i.e. fern spores, mold spores specific to this region and also to oil palm pollen (OP) among the same group of subjects.

MATERIALS AND METHODS

Study population

Eighty-four asthmatic children (55 males and 29 females, age range 2-14 years with a mean age of 7.63 ± 3.31) and seventy-one adult asthmatics (28 males and 43 females, age range 16-80 years with a mean age of 44.56 ± 16.56) were recruited from Pediatric Allergy Clinic and Chest Clinic of the Siriraj Hospital, Mahidol University. Seventy-one healthy adults (28 males and 43 females, age range 20-61 years with a mean age of 23.94 ± 6.54) without any allergic disease were also recruited as controls. Informed consent was obtained from patients or parents prior to entrance into the study.

Skin testing and allergen extracts

Standard SPT²² was used throughout the study. All patients withheld antihistamines for 72 hours (one month for astemizole) prior to skin testing procedure. SPT's were performed on the volar surfaces of

forearms and were read within 15 minutes after the application. A wheal size of 3 mm greater than negative controls (glycerinated saline) was considered positive. Histamine hydrochloride (10 mg/ml) was used as a positive control.

Allergen extracts used in this study were divided into two sets. The first set comprised conventional extracts (Center Laboratories, Port Washington, New York) and consisted of *Dermatophagoides pteronyssinus* (Dp) 10,000 AU/ml, *Dermatophagoides farinae* (Df) 10,000 AU/ml, *Periplaneta americana* (ACR) 1:10 w/v, *Blattella germanica* (GCR) 1:10, w/v, *Alternaria* 1:10 w/v, *Cladosporium* 1:10 w/v, Johnson grass 1:10 w/v, Bermuda grass 1:10 w/v, *Aspergillus* 1:10 w/v, and *Penicillium* 1:10 w/v. The second set comprised extracts prepared locally by the National University of Singapore.²¹ These included extracts of local storage mites, pollens and mold spores which were previously found to be common allergens in Singapore; *Bt*, *Austroglyciphagus malaysiensis* (Am), *Acacia auriculiformis* (Aa), OP, Resam fern spore (RF), *Curvularia fallax* (Cf), and *Exserohilum rostratum* (Er).^{17,21,23,24} The study was reviewed and approved by the Human Rights and Ethic Committee of the Siriraj Hospital.

Statistical analysis

Descriptive statistics were analyzed by SPSS 10.0 statistical package (SPSS, USA). Chi-square and Fisher's exact tests were used to compare positive results to different extracts between adult vs pediatric asthmatics and between adult asthmatics vs controls.

RESULTS

The prevalence of allergen sensitization among the three groups (children, adult asthmatics and controls) were 64.3%, 43.7% and 35.2%, respectively (Table 1). Wheal and flare sizes of histamine controls among children and adults were $5.7 \pm 1.9/24.1 \pm 6.0$ mm and $6.1 \pm 1.9/25.2 \pm 6.3$ mm, respectively. Table 1 demonstrates numbers of patients sensitizing to varying numbers of allergens in each group. Although the sensitization rate among adult asthmatics to any allergen was higher than that of adult controls, the difference was not statistically significant (OR = 1.43, CI = 0.69-2.97, $p = 0.3$).

Table 2 shows numbers and percentages of patients (of the three groups) sensitizing to mites and cockroaches. *Dermatophagoides* were the most common allergens causing sensitization among pediatric

Table 1 Number and percents of sensitization to various allergens among each group of patients (PA = pediatric asthmatics, AA = adult asthmatics, AC = adult controls)

Number of positive skin tests	PA (%)	AA (%)	AC (%)
Any allergens	70 (64.3)	31 (43.7)	25 (35.7)
1 allergen	3 (3.57)	4 (5.60)	3 (4.23)
2 or more allergens	51 (61)	27 (38)	22 (31)

No statistical difference was observed between adult asthmatics and controls.

Table 2 Numbers and percentages of patients sensitized to mites and cockroaches (PA = pediatric asthmatics, AA = adult asthmatics, AC = adult controls)

Allergen	PA (%)	AA (%)	AC (%)	p (OR with CI) AA and AC	p (OR with CI) PA and AA
<i>Dermatophagoides</i>	49 (58.3)	29 (40.8)	23 (32.4)		$p = 0.03$ (2.03, 1.02-4.06)
<i>Dp</i> only	5 (6.0)	2 (2.8)	3 (4.2)	NS	NS
<i>Df</i> only	0 (0)	0 (0)	2 (2.8)		NS
<i>Dp</i> and <i>Df</i>	44 (52.4)	27 (38)	18 (25.4)		NS
Storage mites	24 (28.6)	8 (11.3)	3 (4.2)		$p = 0.008$ (3.15, 1.23-8.32)
<i>Bt</i> alone	8 (9.5)	3 (4.2)	2 (2.8)	NS	NS
<i>Am</i> alone	6 (7.1)	1 (1.4)	0 (0)		NS
<i>Bt</i> and <i>Am</i>	10 (11.9)	4 (5.6)	1 (1.4)		NS
<i>Bt</i> and any <i>Dermatophagoides</i>	18 (21)	7 (9.9)	3 (4.2)	NS	NS
<i>Am</i> and any <i>Dermatophagoides</i>	15 (17.9)	5 (7.0)	1 (1.4)		$p = 0.045$ (2.87, 0.91-9.63)
Cockroaches	23 (27.4)	11 (15.5)	11 (15.5)		NS
American alone	3 (3.6)	4 (5.6)	4 (5.6)	NS	NS
German alone	7 (8.3)	3 (4.2)	2 (2.8)		NS
American and German	13 (15.5)	4 (5.6)	5 (7.0)		NS

NS = No statistically significant

(58.3%) and adult patients (40.8%). The majority of these patients were dually sensitized to *Dp* and *Df*. Of the 49 pediatric patients and the 29 adult patients who were sensitized to *Dermatophagoides*, only 5 children and 2 adults were sensitized to *Dp* alone. Twenty-four children (28.6%) and 8 adult patients (11.3%) were sensitized to storage mites. The rates of sensitization to *Bt* and *Am* were not statistically different from one another among pediatric and adult patients ($p > 0.05$). All pediatric patients sensitized to *Bt* were sensitized to *Dermatophagoides*. Similarly, 15 of the 16 pediatric patients sensitized to *Am*, were also sensitized to *Dermatophagoides* with only one child sensitized to *Am* alone. All adult asthmatics sensitized to these two storage mites were sensitized to the *Dermatophagoides*.

Among 23 children (27.4%) who were sensitized to cockroach allergens, 13 were sensitized to both ACR and GCR whereas only 3 and 7 children were sensitized to ACR or GCR alone, respectively. Of the 11 adult patients (15.5%) sensitized to cockroaches, 4 patients were sensitized to both species while 4 and 3 patients were sensitized to ACR and GCR, respectively.

Table 3 demonstrates percentages of patients sensitized to allergens other than mites and cockroaches. Sensitization to OP among pediatric and adult patients were 8.3% and 5.6% while none of the controls was sensitized to this allergen ($p < 0.05$, controls vs patients, both for adults and pediatric patients). Sensitization rates to other allergens in both patient groups were less than 5%.

A large number of controls were sensitized to the *Dermatophagoides* (32.4%, Table 2). Only 4.2% of controls were sensitized to storage mites. Although sensitization to cockroach allergens among controls was high (15.5%), the rates of sensitization to other pollens and molds were low among this group (Table 3).

DISCUSSION

Previous studies indicated that the rates of allergen sensitization among Thai asthmatic children were between 72 to 93%.^{13,25} The most common allergen causing sensitization among these children were house dust mites.^{13,25,26} Although the two previous reports in a small group of Thai adult asthmatics indicated a range of sensitization to house dust mites to be 76-

Table 3 Numbers and percentages of patients sensitized to allergens other than mites and cockroaches

Allergens	PA (%)	AA (%)	AC (%)
1. <i>Alternaria</i>	4 (4.8)	1 (1.4)	0 (0.0)
2. <i>Cladosporium</i>	4 (4.8)	1 (1.4)	0 (0.0)
3. Johnson grass	2 (2.4)	0 (0.0)	2 (2.8)
4. Bermuda grass	1 (1.2)	2 (2.8)	1 (1.4)
5. <i>Aspergillus</i>	1 (1.2)	2 (2.8)	0 (0.0)
6. <i>Penicillium</i>	0 (0.0)	0 (0.0)	0 (0.0)
7. <i>Acacia auriculiformis</i>	3 (3.6)	2 (2.8)	1 (1.4)
8. Oil palm pollen*	7 (8.3)	4 (5.6)	0 (0.0)
9. Resam fern spore	1 (1.2)	1 (1.4)	0 (0.0)
10. <i>Curvularia fallax</i>	2 (2.4)	0 (0.0)	0 (0.0)
11. <i>Exserohilum rostratum</i>	2 (2.4)	1 (1.4)	0 (0.0)

*Statistically significant comparison between adult asthmatics and controls ($p < 0.05$)

84%,^{27,28} intradermal skin testing was utilized in both reports. This technique could give falsely higher rates of sensitization. Our study is unique in that we studied both pediatric and adult asthmatics and used not only common panel of aeroallergens to Thailand but also newer, important allergens in tropical region such as *Bt*, *Am* and *OP*.^{21,23} The results of our study (Table 1) confirm a high rate of allergen sensitization in asthmatic children (64.3%) and a relatively lower rate in asthmatic adult (43.7%, $p < 0.05$).

Surprisingly, the rate of sensitization among our adult controls is quite high (35.7%) compared to a much lower rate of 20% among healthy controls in Europe (the ECRHS survey - 20% positive to house dust mites,²⁹ 15.8% in New Zealand³⁰ and 17% of Finland).³¹ A large number of our controls were medical students and thus could give high positive rates since it has been shown that atopy is more common in high socioeco-

nomics class.³² However, a similarly high rate of atopic sensitization to house dust mites among healthy Singaporean has been shown recently (34.2 -52.4%).^{21,33}

The most common allergens causing sensitization among asthmatics in our study were the *Dermatophagoides* (Table 2). This finding confirms the results from previous studies from Thailand,^{13,26} Indonesia,¹⁹ Singapore²³ and Malaysia¹⁸ that house dust mites are the most important allergens for Southeast Asia.

Bt, a storage mite from the family *Glycyphagidae*, has been shown to be a major allergen sensitized by patients with asthma and rhinitis residing in tropical regions near the equator, 64% in Brazil,¹⁶ 73% in Taiwan,³⁴ 85% in Colombia¹⁴ and 96% in Singapore.²³ The rates of sensitization to *Bt* of 21.4% in our children and 10% in our adult patients are much lower than in previous reports elsewhere. Most recently, a study from Indonesia using

the same source of *Bt* allergen indicated that the sensitization rates were 75% and 70.6% among pediatric and adult patients residing in Jakarta.¹⁹ The discrepancy between our study and those from other Southeast Asian countries is unclear. In our previous survey of house dust mites fauna in Thailand, *Bt* was rarely observed from dust collected from the Thai homes²⁰ indicating that *Bt* might not be as prevalent in Thailand as in other countries in Southeast Asia.

With the fact that all of our *Bt* sensitized patients were sensitized to the *Dermatophagoides*, cross-reactivity between the two mite species should be considered.^{14,15,35} Indeed, the results of cross enzyme-immunoelectrophoresis indicated that there are at least 4 cross-reactive IgE binding antigens between *Dp* and *Bt*, with *Blo t 5* showing the most cross-reactivity with *Der p 5*.³⁵ However, the results of the studies from Singapore^{35,36} and from RAST inhibitions^{14,15} suggested that *Bt* may possess unique

allergen. Whether *Blomia*-sensitized Thai patients produced specific IgE to only cross-reacting allergens or also to unique *Bt* allergens remains to be clarified.

Cockroaches are another important source of allergen sensitized by atopic patients throughout the world with GCR being the most important allergens sensitized by asthmatic children in inner city USA (36.8%).³ In Brazil, up to 55% of atopic individuals were sensitized to cockroaches.³⁷ In Asia, the rates of sensitization to cockroaches ranged between 40-50%.³⁸ Utilizing intradermal testing, Choovivathana-vanich³⁹ reported a 77% positive rate to mixed cockroach allergens among allergic Thai patients. Our investigation in Thai asthmatic children showed a rate of sensitization to ACR to be 44%.¹³ The sensitization rates of 27.4% and 15.5% among pediatric and adult patients in our study are surprisingly low. Most of our patients were dually sensitized to both ACR and GCR. Nevertheless, the prevalence of cockroach sensitization among asthmatic patient and control is nearly the same as among asthmatics from Norway (9.8% and 7.7%, respectively)⁴⁰ and Korea (11.2%).⁴¹ Recently, Sookrung *et al.*⁴² successfully produced two monoclonal antibodies to major allergens of ACR that are recognized by sera of cockroach sensitive Thai children (Mab 38G6 directed against allergen sizes between 72 to 207 kDa and Mab 3C2 directed against allergen sizes between 45 to 40 kDa). Furthermore, Dirapat *et al.*⁴³ was able to clone the isoallergen of the Per a 1- the major ACR allergen which has been named A6 (LIRSLFGLP amino acid sequence). This allergen bound to

Mab 3C2 and reacted to all serum IgE of the 20 allergic Thai pediatric patients. The new isoallergen of Per a 1 was shown to be a transmembrane protein.⁴³ With the availability of both recombinant allergen and monoclonal antibodies, it is expected that knowledge on allergy to ACR will progress as well as with GCR.

Although the rate of sensitization to OP pollen among our population is relatively low (5% in adult patients and 8% in children), a similar sensitization rate was observed in Indonesia.¹⁹ The rates of sensitization to other pollens and fungal spores among our study population were even lower (< 5%). The similar patterns of sensitization to pollens and spores were shown in the study from Korea,⁴¹ Hong Kong,³⁸ and Malaysia.¹⁸ It is surprising that the rate of sensitization to oil palm pollen among allergic children in Singapore were as high as 40% and with positive rates to fern and mold spores reaching approximately 30%.²¹ This result indicates that pollens and spores are not important sensitizing allergens in Bangkok.

In conclusion, the *Dermaphagoides* are the most important allergens for asthmatics in Thailand. The high rate of sensitization to storage mites among this population could either represent cross-reactivity between domestic and storage mites or could indicate a resurgence of storage mite population within mite fauna in Thailand. Sensitization to pollens and spores are of less importance among this group of patients.

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