

Patterns of Sensitization to Inhalant Allergens among Asthmatic Children in Karbala Province

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Abstract

Background: The patterns of sensitization to inhalant allergens among asthmatic children varies greatly in different countries. The detection of sensitization to inhalant allergens in asthmatic patients is used primarily to establish the diagnosis of allergic asthma. Also it is helpful in reducing asthma exacerbations through allergen exposure avoidance.

Objectives: This study aims to determine the most common inhalant allergens associated with childhood asthma in Karbala province using allergen-specific IgE immunoassay.

Patients and Methods: The present study was conducted in Karbala Teaching Hospital for Children on 75 asthmatic children with elevated total serum IgE levels. All patients were screened for the presence of specific IgE against inhalant allergens most frequently involved in childhood asthma using EUROLINE Pediatric Inhalation IgE test kit that include 20 inhalant allergens. The position of the band was used to determine the specific allergen while the color intensity of the band was used as predictive for the specific IgE concentration in serum samples.

Results: Among 75 patients screened for specific IgE, 60 patients (80%) were sensitized to at least one allergen and allergen-specific IgE was not detected in 15 (20%) patients. 30% of sensitized patients, were sensitized to single allergen and 70% were sensitized to multiple allergens. Cat allergen was found in 60% of sensitized patients followed by European house dust mite allergen (55%). However, dog and aspergillus fumigatus allergens accounted for 45% and 33.3% respectively. Most patients sensitized to cat and dog allergens (55.5% and 52% respectively) showed clear bands signal intensity (moderate allergen-specific IgE concentration). However the majority of patients sensitized to other inhalant allergens showed weak bands signal intensity (low allergen-specific IgE concentration).

Conclusions: Animal allergens (mostly cat) are the predominant inhalant allergens associated with childhood asthma in Karbala province with relatively higher allergen-specific IgE concentrations than house dust mite and fungal allergens.

Key words: Childhood asthma, Inhalant allergens, Specific IgE, Karbala.

الخلاصة

تختلف أنماط التحسس للمحسسات المستنشقة بين الأطفال المصابين بالربو اختلافا كبيرا في مختلف البلدان. إن الكشف عن التحسس للمحسسات المستنشقة في مرضى الربو يستخدم أساسا في تشخيص الربو التحسسي. كما أنه مفيد في الحد من تفاقم مرض الربو من خلال تجنب التعرض لمسببات الحساسية. تهدف هذه الدراسة إلى تحديد المحسسات المستنشقة الأكثر شيوعا عند الأطفال المصابين بالربو في محافظة كربلاء باستخدام تقنية المقاييس المناعية لقياس الغلوبولين المناعي نوع E المحدد للمحسسات المستنشقة. أجريت هذه الدراسة في مستشفى كربلاء التعليمي للأطفال على 75 طفلا مصابين بالربو. تم فحص جميع المرضى لوجود الغلوبولين المناعي نوع E المحدد بالمحسسات المستنشقة. اشتمل المسح على 20 محسس مستنشق مرتبط عادة بربو الأطفال. تم استخدام موقع الحزمة لتحديد نوع المحسس المستنشق في حين تم استخدام كثافة اللون في الحزمة كمقياس لتركيز الغلوبولين المناعي نوع E المحدد في عينات مصل الدم. أظهرت الدراسة أن 80% من

المرضى كانوا متحسسين لمحسس مستنشق واحد على الأقل ولم يتم الكشف عن أي محسس في 20% من المرضى. كان التحسس لمحسس مستنشق واحد موجود في 30% من المرضى المتحسسين والتحسس لعدة محسسات مستنشقة موجود في 70% من المرضى المتحسسين. معظم التحسس كان للقطط (60% من المرضى) يليه التحسس لعث غبار المنزل الأوربي (55%) ثم التحسس للكلاب والرشاشيات الدخناء (45% و 33.3% على التوالي). في أغلب المرضى المتحسسين للمحسسات المستنشقة الخاصة بالقطط و الكلاب (55.5% و 52% على التوالي) كان الغلوبولين المناعي نوع E المحدد متوسط التركيز، بينما كان التركيز المصلي منخفض لباقي المحسسات المستنشقة. نستنتج من هذه الدراسة بأن المحسسات المستنشقة الحيوانية (وخصوصا القطط) هي المحسسات الأكثر شيوعا بين الأطفال المصابين بالربو في محافظة كربلاء وإن التركيز المصلي للغلوبولين المناعي نوع E المحدد لهذه المحسسات هو أعلى نسبيا من باقي المحسسات كعث غبار المنزل و المحسسات الفطرية.

Introduction

Asthma is the most common chronic disease among children in different countries [1-3]. There are two main phenotypes of persistent childhood asthma; *Atopic* phenotype (allergic or atopic asthma) and *non-atopic* phenotype (non-allergic or non-atopic asthma) [4]. Allergic asthmatic children often continue to have the disease in adulthood [5]. The diagnosis of allergic asthma requires the presence of sensitization to an inhalant allergen documented by positive allergen-specific IgE test or skin prick test [6]. Total serum IgE (TSIgE) level can be used to differentiate between atopic and nonatopic asthmatics prior to allergen-specific IgE determination [7]. The patterns of sensitization to inhalant allergens among asthmatic children varies greatly in different countries. Also there is variability between different regions within the same country [2]. Indoor inhalant allergens mostly involved in the development of asthma include house dust mites (HDM), animal proteins (particularly cat and dog allergens) and fungi [8]. The exposure to outdoor inhalant allergens like pollens of trees, grasses and weeds has less significant role in the development of asthma, but usually associated with the precipitation of asthma attacks [9]. The detection of sensitization to inhalant allergens in asthmatic children is used primarily to establish the diagnosis of allergic asthma [6]. Also it is helpful in reducing asthma exacerbations through avoidance of exposure to particular allergen and considering allergen immunotherapy (desensitization) [1].

Objectives: This study aims to determine the most common inhalant allergens associated with childhood asthma in Karbala province using allergen-specific IgE immunoassay.

Patients and Methods

The present study was conducted in Karbala Teaching Hospital for Children on 75 asthmatic children consulted the outpatient clinic of the hospital in the period from June 2011 to June 2012. Patients with at least 3 attacks of reversible bronchoconstriction in the past 12 months [10] and elevated TSIgE (documented by positive IgE screening test) were included in the study.

Venous blood was obtained from each patient and allowed to clot then serum was separated by centrifugation. IgE Serum RapiCard InstaTest (Cortez, USA, LOT No. 1102081) was used as a screening test for elevated TSIgE. The test detected TSIgE levels above 80 IU/ml (cutoff value). All patients included in the study were screened for the presence specific IgE against different inhalation allergens. For this purpose, EUROLINE Pediatric Inhalation IgE test kit (Euroimmune, Germany, LOT No. A100728AA and A120111AB) was used which is semiquantitative immunoassay test that contains test strips coated with 20 different inhalant allergen extracts that most frequently involved in childhood asthma [11]. The test was considered as positive, when a band signal appears in the position of the specific allergen while the color intensity of the band was used as predictive for the specific IgE

concentration in serum sample. Table -1- divided the signal into 4 classes which correspond to the band. The classification was done by comparing the color intensity of the band with that of the indicator band of the same strip (figure -1-).

For patients showed negative results with Pediatric Inhalation IgE test kit, another screening for the presence specific

IgE against inhalation allergens was done using, EUROLINE Inhalation Gulf IgE test kit (Euroimmune, Germany, LOT No. A120411AC) that contains test strips coated with 20 different inhalant allergen extracts (6 were new allergens and not included in the Pediatric Inhalation IgE test kit) most frequently involved in asthmatic patients of the Arab Gulf region.

Table 1: Interpretation of allergen-specific IgE results according to band intensity

Class	Result	Characteristics
0	Negative (Allergen-specific IgE concentration < 3.5 IU/ml)	No band
+	Low allergen-specific IgE concentration (3.5-17.5 IU/ml)	Weak band signal
++	Moderate allergen-specific IgE concentration (17.5-50 IU/ml)	Clear band signal
+++	High allergen-specific IgE concentration (> 50 IU/ml)	Intense band signal

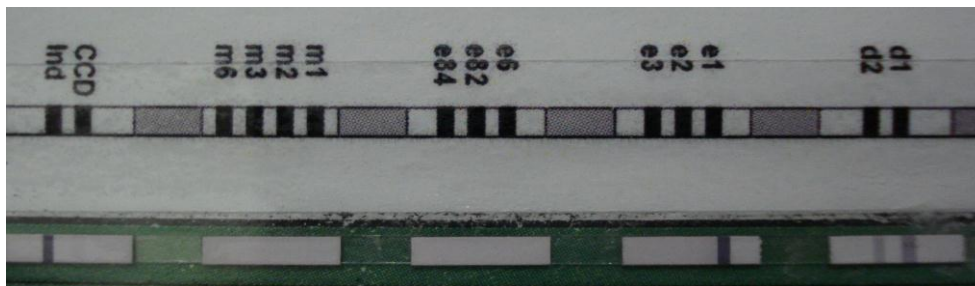


Figure 1: EUROLINE Pediatric Inhalation (IgE) test strip for an asthmatic child showing intense band signal (+++) at (e1) allergen, clear band signal (++) at (d1), weak band signal (+) at (d2) and no band (0) at (e2, e3, e6, e82, e84, m1, m2, m3 and m6). An intense indicator band also shown.

Results

The study included 75 asthmatic children with elevated TSIgE levels aged from 2-13 years (mean ± SD = 6.53±3.18 years), 49 (65.3%) were males and 26 (34.7%) were females (male to female ratio = 1.9 : 1) . 20 (26.7%) patients were from rural areas and 55 (73.3%) patients were from urban areas. All patients were screened for specific IgE against pediatric inhalant allergens, 60 (80%) patients were sensitized (showed positive results) to at least one allergen. Allergen-specific IgE was not detected in 15 (20%) patients. Table -2- showed that 18 patients (30%) were sensitized to single allergen and 42

patients (70%) were sensitized to multiple allergens.

Cat allergen (e1) was found in 60% of sensitized patients followed by dermatophagoides pteronyssinus mite (European HDM) allergen (55%). However, dog and aspergillus fumigatus allergens accounted for 45% and 33.3% respectively (table -3-). The majority of patients sensitized to two allergens showed positive results to (European HDM + aspergillus fumigatus) allergens (33.3%) followed by (cat + dog) allergens (26.6%). Most patients (37.5%) sensitized to three allergens showed positive results to (cat + dog + European HDM) allergens, while the majority of patients (50%) sensitized to

four allergens showed positive results to (cat + dog + European HDM + aspergillus fumigatus) allergens.

Regarding the band intensity (which reflects the relative concentration of allergen-specific IgE), table 4- demonstrated that 57.9% of the bands were of weak (low allergen-specific IgE concentration) band signals, 38.5% of clear (moderate concentration) band signal and 3.6% of intense (high concentration)

band signals. Cat allergens were the predominant allergens showed intense and clear band signals. Most patients sensitized to cat and dog allergens (55.5% and 52% respectively) had clear bands signal intensity (moderate specific IgE concentration). However the majority of patients sensitized to other inhalant allergens had weak bands signal intensity (low specific IgE concentration) (table 4-).

Table 2: Frequency of specific IgE against pediatric inhalation allergens in sensitized asthmatic children according to allergens numbers

Number of allergens	Number of sensitized patients	Percentage of sensitized patients
1	18	30%
2	15	25%
3	8	13.3%
4	8	13.3%
5 and above	11	18.4%
Total	60	100%

Table 3: Frequency of specific IgE against pediatric inhalation allergens in sensitized asthmatic children according to allergen type.

Code	Allergen type	Number and percentage of sensitized patients (N=60)
e1	Cat	36 (60%)
d1	Dermatophagoides pteronyssinus (European HDM)	33 (55%)
e2	Dog	27 (45%)
m3	Aspergillus fumigatus	20 (33.3%)
m6	Alternaria alternate	11 (18.3%)
e84	Hamster	10 (16.7%)
e3	Horse	7 (11.7%)
m2	Cladosporium herbarum	7 (11.7%)
w9	Plantain	7 (11.7%)
e82	Rabbit	6 (10%)
d2	Dermatophagoides farinae (American HDM)	5 (8.3%)
w6	Mugwort	5 (8.3%)
g6	Timothy grass	4 (6.7%)
g12	Cultivated rye	4 (6.7%)
t2	Alder	4 (6.7%)
t3	Birch	4 (6.7%)
e6	Guinea pig	2 (3.3%)
m1	Penicillium notatum	2 (3.3%)
t4	Hazel	1 (1.6%)
w8	Dandelion	0 (0%)

From the 15 asthmatic children showed negative results to pediatric inhalation allergens, 10 patients showed positive results to at least one allergen with Gulf inhalation allergens and allergen-specific IgE was not detected in 5 patients. Two patients showed positive results for cockroach and olive, two patients for

cockroach alone, two patients for camel, two patients for oak and two patients for candida albicans allergens. Thereby, the total number of sensitized patients became 70 (93.3%) out of 75 asthmatic children screened with 26 different inhalant allergens, and only 5 (6.7%) patients showed negative results.

Table 4: Frequency of specific IgE against pediatric inhalation allergens in sensitized asthmatic children according to allergen type and band intensity.

Code	Allergen type	Bands signal intensity			Total
		Intense No. (%)	Clear No. (%)	Weak No. (%)	
e1	Cat	3 (8.5%)	20 (55.5%)	13 (36%)	36 (100%)
d1	Dermatophagoides pteronyssinus (European HDM)	1 (3%)	11 (33%)	21 (64%)	33 (100%)
e2	Dog	-	14 (52%)	13 (48%)	27 (100%)
m3	Aspergillus fumigatus	1 (5%)	5 (25%)	14 (70%)	20 (100%)
m6	Alternaria alternate	1 (9%)	5 (45.5%)	5 (45.5%)	11 (100%)
e84	Hamster	-	5 (50%)	5 (50%)	10 (100%)
e3	Horse	-	1 (14%)	6 (86%)	7 (100%)
m2	Cladosporium herbarum	-	1 (14%)	6 (86%)	7 (100%)
w9	Plantain	-	4 (57%)	3 (43%)	7 (100%)
e82	Rabbit	-	-	6 (100%)	6 (100%)
d2	Dermatophagoides farinae (American HDM)	1 (20%)	2 (40%)	2 (40%)	5 (100%)
w6	Mugwort	-	2 (40%)	3 (60%)	5 (100%)
g6	Timothy grass	-	2 (50%)	2 (50%)	4 (100%)
g12	Cultivated rye	-	2 (50%)	2 (50%)	4 (100%)
t2	Alder	-	-	4 (100%)	4 (100%)
t3	Birch	-	1 (25%)	3 (75%)	4 (100%)
e6	Guinea pig	-	-	2 (100%)	2 (100%)
m1	Penicillium notatum	-	-	2 (100%)	2 (100%)
t4	Hazel	-	-	1 (100%)	1 (100%)
w8	Dandelion	-	-	-	-
	Total	7 (3.6%)	75 (38.5%)	113 (57.9%)	195 (100%)

Discussion

The exposure of asthmatic children to inhalant allergens has been shown to increase airway inflammation, airway hyperresponsiveness, asthma symptoms, the needs for medication, and death due to asthma [12]. Diagnosis of sensitization to inhalant allergens by skin prick test has limitations, like limited number of allergens that can be tested and the risk of

development of allergic reactions [13]. Therefore there is increasing role for allergen-specific IgE immunoassay as screening test for sensitization [14]. In this sense, several panels of inhalant allergen-specific IgE tests have been used for screening patients with respiratory diseases in different parts of the world [11,14].

In the present study, screening of asthmatic children for the presence of specific IgE against pediatric inhalant

allergens demonstrated that sensitization to multiple inhalant allergens was the predominant pattern of sensitization (70% of patients). Closely similar finding (67.4% of patients sensitized to multiple allergens) was reported by Moheeb Salih in Tikrit province who studied asthmatic children and adults using skin prick test [15].

This study showed that cat allergen was the predominant allergen associated with childhood asthma (detected in 60% of patients) followed by European HDM (55%), dog (45%) and aspergillus fumigatus (33.3%) allergens. Studies in other parts of Iraq on asthmatic patients (children and adults) using skin prick test showed that HDM followed by fungal allergens were the most common allergens in both Tikrit [15] and Diyala [16]. This difference in distribution of common allergens is partly due to geographical variation and the fact that both studies screened adults and children. The sensitization to domestic animals is generally more common among children [17]. The pattern of allergens involved in multiple sensitization presented by this study (animals + HDM + fungal) is generally consistent with Salih study [15].

In sensitized asthmatic children the severity of asthma symptoms is strongly correlated with serum levels of allergen-specific IgE, with higher serum levels associated with more severe symptoms [18]. One of the major advantages of allergen-specific IgE immunoassay over skin prick test is the determination of specific IgE concentration [7].

The present study demonstrated that cat allergens were the predominant allergens showed high and moderate serum allergen-specific IgE concentrations and patients sensitized to cat and dog allergens have relatively higher concentrations of allergen-specific IgE than those sensitized to both HDM and fungal allergen. These findings increased the clinical relevance of sensitization to domestic animals in Kerbala province.

An important aspect in allergen-specific IgE immunoassay is that, the allergens present in the test kit should represent the most common allergens involved in that part of the world. This required multiple screening studies in different geographical areas to determine the allergen panel for particular disease [14].

The results of this study showed that 10 patients (out of 15 asthmatic children showed negative results to pediatric inhalation allergens) showed positive results to at least one allergen with Gulf inhalation allergens (allergen-specific IgE test kit frequently used to screen asthmatic adults in the Arab Gulf region). The new allergens were: cockroach, camel, olive, oak and candida albicans allergens.

Conclusions

Animal allergens (mostly cat) are the predominant inhalant allergens associated with childhood asthma in Karbala province with relatively higher allergen-specific IgE concentrations than house dust mite and fungal allergens.

Recommendations

Using EUROLINE Pediatric Inhalation IgE test kit as a screening test for sensitization among asthmatic children in Iraq, may miss some sensitized patients. Therefore, it is recommended to develop a special Iraqi panel of most common inhalant allergens associated with childhood asthma in the future, taking in consideration the results of this study and other similar Iraqi studies.

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