Significance of IgG_4 Subclass in Atopic Dermatitis: Comparison with total and specific IgE

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Abstract:

Objective: To assess the clinical significance of estimating total IgG, IgG_4, subclass and specific IgE testing to some food and aeroallergens in atopic dermatitis.

Patients and Methods: Twenty seven atopic dermatitis patients (11 infants and 16 children) were subjected to detailed medical history and clinical evaluation. They were also assessed immunologically via estimation of total IgE, IgG, IgG_4, and specific IgE to some food and aeroallergens.

Results: Raised serum IgE was detected in 70.4% of our patients and this increase was related to the severity of the eczema. One third of our series was sensitized to some food and aeroallergens. Sensitization to mite antigen was detected in 5 of the 6 asthmatic patients. The levels of total serum IgE were significantly high in both asthmatic and mite sensitive patients.

Conclusion: The complex of pathogenesis of atopic dermatitis makes it difficult to judge the relative importance of raised IgE and IgG_4 in modulating the disease.

Key words: Atopic dermatitis, IgG4, total IgE, specific IgE

Introduction:

Atopic dermatitis (AD) is a genetically determined cutaneous inflammatory condition associated with numerous pharmacological and immunological abnormalities as well as a tendency to produce specific IgE (reagins) in response to common environmental antigens. AD is a major cause of morbidity in children. It affects up to 20% of population.

The disease lacks a primary skin lesion, furthermore, there are no specific diagnostic histological features or characteristic laboratory markers.(1) However, Hanifin and Rajka distinguished 4 major and 23 minor features of AD.(2) The diagnosis of AD needs at least 3 major and 3 minor features.

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The fundamental defects in AD remain largely unclear. It is a disease with a multifactorial etiology. Hereditary background, immunological abnormalities, climatological circumstances, emotional and physical stress as well as cutaneous infections are the main etiologic factors in its pathogenesis.(3)

The immune system is involved in the pathogenesis through lymphocyte mediated inflammation of skin and increased incidence of type I allergies induced by environmental allergens. Food and aeroallergens may provoke lesions and have been implicated as etiologic agents in some cases.(4)

Several authors have suggested that reaginic antibodies not belonging to the IgE class occur in man. Such antibodies could possibly belong to subclass IgG4.(5-9)

Thus, the aim of our study is to clarify the clinical significance of estimating total IgG and IgG_4 subclass, as well as to study the importance of specific IgE testing to some food and aeroallergens in AD.

Patients and Methods:

The study was performed on 27 Kuwaiti patients whose skin lesions satisfied the criteria of AD as defined by Hanifin and Rajka.(2) They were selected from the outpatient clinic of Dermatology department, Salmiyah clinic, State of Kuwait.

Severity of the disease was graded as mild, moderate or severe based on extent, course and intensity of eczema.(10) Body surface involvement was estimated by the rule of nines.(11)

According to the age of our patients, they were divided into 2 groups (infantile AD < 2 years old and childhood AD > 2 years up to 14 years old).

Ten subjects (non-atopic volunteers) were selected as controls in the present study. They were dermatologically free, non asthmatic and had no history of allergic rhinitis.

All patients and controls were subjected to:

A. Detailed medical history & Clinical Evaluation

B. Laboratory assessment via:

1) Complete blood picture, urine and stool analysis
2) Immunological studies :-

One) Determination of total serum IgG and IgG4 subclass using rate nephelometry method (Beckman assay protein system 015-248412-A; USA).

Two) Estimation of total serum IgE using Sandwich ELISA; (Biomerieux; VIDAS total IgE, 30419).

Three) Testing for specific IgE to some food and aeroallergens using ELISA (Pasteur CERBA Laboratory Paris).

Reference Ranges:
Normal ranges of total serum IgG and IgG4 subclass levels are: 2.3-15.4 g/L and 0.01-1.64 g/L respectively.

Reference ranges for total serum IgE levels differ according to age. Normal ranges are <30 ku/L, <40 ku/L and <118 ku/L for age groups < 5-< 10 years, 10 years and over respectively.

Results of specific IgE are scored as uninterpretable, weak, moderate, strong, very strong and graded accordingly from 0 to 4 classes. For practical reasons we considered class 2 (moderate), class 3 (strong) and class 4 (very strong) as a positive result.

Results:
Our cohort was 27 atopic dermatitis patients; 11 infants and 16 children. Detailed clinical data, major and minor features of AD observed in our patients are presented in Table I and II.

Moderate to severe degrees of AD were seen in 16 patients (59.3%) and the extension of the body surface involvement ranged from 21-95% (x=63.8%).

Peripheral blood eosinophilia (>760cells/μL) was seen in 6 patients (22.2%); 2 infants and 4 children.

Results of total serum IgE, IgG and IgG_4 subclass levels in infantile and childhood AD are summarized in Table III and IV. There was no significant difference in the mean levels of these Igs in studied infantile and childhood groups.

Raised total serum IgE was detected in 8 of 11 infants (72.7%) and 11 of 16 children (68.8%) i.e. 19(70.4%) of our patients had high total serum IgE levels. Total serum IgE levels were significantly higher (X=2861.4 Ku/L) in patients with moderate to severe AD than in those with mild form (X=347.9 Ku/L); P<0.005. Total serum IgG was raised in 2 patients (7.4%), High IgG_4 levels were seen in 7 patients (25.9%); 3 infants and 4 children.

A comparison between IgG_4 and IgE levels in our atopic patients showed four groups of patients: high in both IgE and IgG_4 levels (No=4, 14.8%), low IgE and IgG4 levels (No=5, 18.5%), high IgE and low in IgG_4 (No=15,55.6%) and low in IgE and high in IgG_4 (No=3, 11.1%).

There was statistically insignificant negative correlation between total serum IgE levels and IgG4, values (r=0.113, P>0.0005).

Results of specific IgE antibody determination against some food and aeroallergens in infantile and childhood AD are shown in Table V and VI. Specific IgE antibodies against food allergens were detected in 9 patients (33.3%). Six of whom showed sensitization to both food and aeroallergens and the other three patients had sensitivity to only some food or aeroallergens. Eggs, milk and fish were the most frequent food allergens detected while Dermatophagoide Pteronyssinus (DP) was the commonest aeroallergen observed.

Six of our atopic patients were asthmatic (22.2%); 5 of whom were sensitized to DP mite antigen. The onset of dermatitis was below the age of 6 months in 4 of those asthmatic patients. The levels of total IgE were significantly high in asthmatic and mite sensitive patients; Table VII.

Nineteen of our cohort (70.4%) had positive history of food intolerance (8 infants and 11 children). There was no statistically significant relation between the history of food intolerance and the high levels of total serum IgE, IgG and IgG4, as well as the specific IgE antibody results to food allergens Table VIII.

Two young children had positive specific IgE against maize antigen although their mothers assumed that they had never eaten it. Our control subjects were 3 infants (mean age-18.4 months) and 7 children (mean age-11.2 years). No abnormal laboratory findings could be detected in the control subjects; except mild increase of serum IgE in only one of them (140 K μ/L).

Table I :
Major and minor features of AD seen in our patients

Features	Group I Infantile AD	Group II Childhood AD
Major Features	*No. (%)*	*No (%)*
(1) Pruritus	11 (100)	16 (100)
(2) Chronic or Chronic relapsing Dermatitis	1 (100)	16 (100)
(3) Typical clinical feature	9 (81.8)	14 (87.5)
(4) Personal/Family history (PH/FH) of AD or related atopic disease	(PH2) (18.2) (PH5) (31.3)	(FH7) (63.4) (FH 9) (56.3)
Minor features		
(1) High serum IgE	8 (72.8)	11 (68.8)
(2) Food intolerenace	8 (72.8)	11 (68.8)
(3) Xerosis	7 (63.6)	13 (81.3)
(4) White dermographism	8 (72.8)	12 (75)
(5 Dennie morgan infraorbital fold	1 (9.1)	3 (18.8)
(6) Intolerance to wool	-	4 (25)
(7) Influence of emotions	-	5 (31.5)
(8) Tendency to skin infection	3 (27.3)	7 (43.8)
(9) Cheilitis	-	3 (18.8)
(10) Periobital darkening	-	3 (18.8)
(11) Keratosis pilaris	5 (45.5)	9 (56.3)
(12) Nipple eczema	-	2 (12.5)
(13) Pityriasis alba	2 (18.2)	4 (25)
(14) Hand/foot eczema	6 (54.6)	10(62.5)

Table II:
Clinical profile and type of eczema in infantile and childhood AD

	Infantile AD (No.=11)	Childhood AD (No.=16)
Age in years		
X (average)	1.4	8.4
Range	0.8-2	3.5-13.5
Age of onset		
X	7.5 months	9.5 months
Sex		
M:F	6:5	9:7
Site affected		
Face	9 (81.8%)	9 (56.3%)
Flexors (F)	4 (36.4%)	10 (62.5%)
Extensors (E)	1 (9.1%)	6 (37.5%)
(F) + (E)	0	2 (12.5%)
Morphological type of eczema		
Acute	7 (63.6%)	4 (25)
Subacute	4 (36.4%)	10 (62.5%)
Chronic	-	2 (12.5%)
Grades of AD		
Mild	6 (54.5%)	5 (31.2%)
Moderate	4 (36.4%)	7 (43.8%)
Severe	1 (9.1%)	4 (25%)

Table III:
Levels of IgE, IgG and IgG4 subclass in infantile and childhood atopic dermatitis

Ig	Group	No	Range	X	Sd	Ttest
Total IgE	Infantile	11	17.1-335	122.8	99.34	t=1.424 NS
(Ku/L)	Childhood	16	20.4-20000	2329.4	5107.58	P>0.05
Total IgG	Infantile	11	9.2-16.7	12.7	2.27	T=1.156 NS
(g/L)	Childhood	16	5.9-17.1	13/3	3.53	p>0.05
IgG4	Infantile	11	0.2-2.6	1.3	0.7	t=0.663 NS
(g/L)	Childhood	16	0.3-2.8	1.1	0.87	p>0.05

Table IV:
High levels of total serumf IgE, IgG and IgG_4 Subclass in infantile and childhood atopic dermatitis

Ig	Group	No	%	Range	X	Sd	Ttest
High total	Infantile	8	72.7	84.4-335	161.6	88.1	
IgE (Ku/L)	Childhood	11	68.8	195-20000	3367.4	5944.6	NS
	Total	19	70.4	84.4-20000	2017.1	4720.17	
High total	Infantile	1	9.1	-	16.7	-	
IgG g/L	Childhood	1	6.3	-	17.1	-	NS
	Total	2	7.4	16.7-17.1	16.9	0.28	
High IgG_4	Infantile	3	27.3	1.9-2.6	2.2	0.35	
g/L	Childhood	4	25	1.8-2.8	2.3	0.49	NS
	Total	7	25.9	1.8-2.8	2.3	0.482	

X = average Sd = standard of deviation,
Ns = not significant T = Tyros

Table V:
Specific IgE to some food allergens in infantile and childhood AD

Food allergens	Infantile AD Total No. = 11 Positive	Childhood AD Total No. =16 Positive	Total No 27 Positive
Nuts	-	2	2
Tropical fruits	2 (mixed)	-	2
Meat	-	-	-
Sea products	1 (fish)	2 (fish)	3
Flours	-	2 (wheat, rice, maize)	2
Dairy products	2 (mild)	2 (milk)	4
Egg	2 (1 yolk, 2 white)	3 (1yolk, 3 white)	5
Total No.	* 4 (36.4%)	** 5 (31.2%)	9 (33.3)

* Three patients were sensitive to more than one food allergens
* Four patients were sensitive to more than one food allergens

Discussion:

It is generally agreed that the reported prevalence of AD has been increasing over the last 30 years [12,13]. AD represents the most frequent dermatitis among children in Kuwait (31.3%).[14]

In the present study, 27 infants and children were diagnosed to have AD. The frequency of major and minor clinical features in those patients corroborate other studies. [15-19] Twenty two percent of our series had peripheral blood eosinophilia which is almost identical to that obtained by Nancy et al[20]. It has been reported that in AD, the number of eosinophils and the quality of their products, cationic protein and major basic protein, are increased in peripheral blood and the increase correlates with the clinical severity of the skin disease.[3, 20]

Many investigators have found raised IgE in approximately 70% of patients with AD. This increase appears to be related to the severity and extent of the disease.[1,3,4,10,21] These findings

Table VI:
Specific IgE to some aeroallergens in infantile and childhood AD

Aeroallergens	Infantile AD Total No. = 11 Positive	Childhood AD Total No. =16 Positive	Total No 27 Positive
Tree pollens	-	-	-
Weed Pollens	-	-	-
Grass pollens	-	-	-
Animal dander	1 (dog dander)	3 (dog, cat, horse)	4
Moulds	-	2 (candida)	2
Insects	-	1 (cockroach)	1
Mites			
DP	2	6	8
D Farinae	-	-	-
Total No.	3 (27.3%)	* 6 (37.6%)	9 (33.3%)

* Four patients were sensitive to more than one aero allergen

Table VII:
Total serum IgE levels in AD patients in relation to bronchial asthma and sensitivity to mite antigen

Total IgE		No.	X	SD	T test
No=27	Asthmatic	*6 (22.2%)	5283	7754.7	t=9.39
	Non-asthmatic	21 (77.8%)	329.7	631.5	p>0.001 (sig)
No=27	+ve mite sensitivity	8 ** (29.6%)	3499.8	6735.9	t=2.367
	-ve mite sensitivity	19 (70.4%)	822.3	2113.9	p<0.05 (sig)

* Five of them were sensitive to mites
* All had high IgE and 5 of them were asthmatic

Table VIII:
Total serum IgE, specific IgE to food allergens and IgG4 subclass in AD: their relation to history of food intolerance

	History of Food intolerance + ve	- ve	Total
Total IgE: No. (%)			
High Level	14 (73.7)	5 (62.5%)	19 (70.4)
Normal Level	5 (26.3)	3 (37.5%)	8 (29.6%)
Total P>0.05 (NS)	19 (100%)	8 (100%)	27 (100%)
Specific IgE to food allergens			
+ ve	5 (26.3)	4 (50%)	9 (33.3)
- ve	14 (73.7)	4 (50%)	18 (66.7)
Total P>0.05 (NS)	19 (100%)	8 (100%)	27 (100%)
IgG_4			
High level	5 (26.3)	2 (25%)	7 (25.9)
Normal level	14 (73.7)	6 (75%)	20 (74.1)
Total P>0.05 (NS)	19 (100%)	8 (100%)	27 (100%)

NS : Not significant

confirm our results as 70.4% of our patients were found to have high total serum IgE. The levels were significantly higher in moderate to severe AD than in mild form. Normal persons may have mild increase of serum IgE.[3] The same was detected in a single control subject in the present work.

There has been considerable controversy concerning the possible pathogenetic role of various IgG subclasses in the pathogenesis of allergic disease. IgG_4 represents only 3-4% of total IgG and it has been found to be increased in AD and asthma.[22-26] In our study, 7 patients (25.9%) had raised serum IgG_4 levels. Four of them had also high serum IgE while the other 3 patients had normal IgE levels and history of food intolerance. The latter group is of particular interest and suggested that a role of IgG4 exists in those patients as well as specific IgG_4 against food allergens is recommended to be done.

In our cohort, there was statistically insignificant negative correlation between total serum IgE and IgG_4 levels. This begs the question, how IgG_4 and IgE coregulated. Until now, it is not known that B cell has, at some point, a choice between the two IL4 dependent isotypes, IgE and IgG_4. [27,28]

The high serum levels of allergen-specific IgE suggest a role for IgE in the mechanism involved in allergic inflammation in patients with AD. The route by which allergens influence the course of AD may be the respiratory tract, gastrointestinal tract or the skin.[21] The controversial question of the role of food in causing or exacertabing lesions of AD has not been fully answered.[29] One third of our patients had raised specific IgE against some food allergens. In the literature, the frequency of a clinically important food allergy in children with AD has ranged from 33%, 39% to 56%.[30] The most common allergenic food, worldwide, are eggs, milk, fish, peanut and wheat.[31] Our study supports these findings where high specific IgE antibodies against egg, milk, fish were the most frequently encountered.

In the present series, there was insignificant relation between the history of food intolerance and the levels of immunoglobulins measured as well as specific IgE against food allergens. Furthermore, two of our patients had positive specific IgE tests for foods they had never eaten.

This could be explained because a molecule against an epitope

on one food will also recognize similar epitope on related food. However it is possible that they had been exposed to these allergens unknowingly, eg. in commercially prepared foodstuff. So, some authors believe that the only accepted method for establishing a firm diagnosis of food hypersensitivity is a double blind placebo controlled food challenge or by open, controlled challenge. [32,33]

We found a clear correlation between high serum IgE levels and the associated asthma in our patients with AD which has also been confirmed by others. In addition, asthma was more frequent in our AD patients with onset of skin lesions before the age of 6 months (4 of the 6 asthmatic patients).[1,3,15,17]

The role of aeroallergens (specially mites) in the pathophysiology of inflammatory lesions of AD has been demonstrated in recent studies by means of patch test and skin biopsy. Thus children suffering from AD, presenting an epidermis with an altered barrier, have enhanced sensitization by airborne allergens which cause eczematous lesions. [34,35] Sensitization to mite antigens has been reported to occur in 20-60% of AD patients. Furthermore, 80% of allergic subjects were proved to have circulating antibodies against dermatophagoid antigens as well as they probably are the main triggers of asthma.[36,37] In our patients, sensitization to DP mite was detected in 8 (29.6%); 5 of whom were asthmatic.

Conclusions:

The complexity of pathogenesis of AD makes it difficult to judge the relative importance of raised IgE and IgG4 in modulating the disease. A group of patients who has high IgG_4 and low IgE needs further assessment to clarify the role of IgG4 antibody in them.

The diagnosis of food allergy in AD is difficult and poor correlation between clinical history and specific immunologic test prevails. So, all positive specific IgE results to food, must be evaluated along with clinical history and oral challenge to arrive at accurate diagnosis. Patients having AD associated with bronchial asthma should attract the attention of dermatologist to seek out house dust mite as a possible triggering or exacerbating factor.

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