

An Evaluation of Cockroach Allergies in Atopic Dermatitis

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Background: Atopic dermatitis (AD) is primarily influenced by environmental factors including exposure to pollutants and indoor allergens (particularly, house dust mites). Although house dust mite antigens are the most prevalent components of indoor allergens in Korea, cockroaches also can be considered to act as an important allergen.

Object: This study was done to evaluate the differences in three different atopic patch test (APT) techniques, and the relationship between APT and skin prick test, total IgE, and specific serum IgE level using cockroach allergen.

Methods: We performed patch test in 57 patients with AD and 30 normal controls on clinically lesional and normal appearing skin with German cockroach (GC) allergens (extract, as is) in three different techniques (standard, scratch, DMSO). Reactions were evaluated after 48 hours, and compared with the results of skin prick test, total and specific IgE levels. Detailed atopy history and severity scoring were taken.

Results: In the GC (whole body) prick test positive group, there was 1263.02 IU/ml of total IgE, and this amount was significantly higher than GC (whole body) prick test negative group who had 549.46 IU/ml ($p < 0.05$). The positive reaction rate to whole body of American cockroach (AC) was significantly higher in the patient group than control group ($p < 0.05$), but the positive rate to whole body of GC was high, but not significantly high in the patient group ($p = 0.053$). There were significant differences in positive patch test reactions to either Ext or As is between patient group and control group ($p < 0.05$). The positive rate to As is was significantly higher than to Ext in either lesional skin or non-lesional skin in patient group ($p < 0.05$). But there were no considerable differences in positive reactions to either Ext or As is between 3 different methods (standard, scratch, DMSO mix) in both lesional and non-lesional areas ($p > 0.05$). There was no significant relationship between the positive reactions to patch test and prick test to Ext and As is antigen. The APT results showed no significant concordances with skin prick test and RAST for cockroach antigens.

Conclusions: APT seems to be a different dimension of atopic skin inflammation and may provide further diagnostic information in addition to a patient's history, skin prick test, and RAST results. (*Ann Dermatol* 15(2) 52~59, 2003).

Key Words : Atopic dermatitis, Atopy patch test, Cockroach allergen

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Atopic dermatitis (AD) is a chronically relapsing skin disorder that occurs most commonly in early infancy and childhood. It has been suggested that AD occurrence is primarily influenced by environmental factors including exposure to pollutants and indoor allergens (particularly, house dust mites)¹. Two types of immunologic reactions, im-

mediate-type and delayed-type, can occur after application of aeroallergens in AD patients. The immediate-type of reaction is seen right after subcutaneous application of aeroallergens like house dust mites. Delayed-type eczematous skin reactions appear in 24 to 48 hours after the epicutaneous application of aeroallergens.

It is not clear if sensitization to inhaled allergens in AD is caused by exposure in the airways or to the skin². Most previous studies on atopic patch test (APT) were done using only mite antigens, and with grass pollen and birch pollen in some studies. Although house dust mite antigens are the most prevalent components of indoor allergens in Korea, cockroaches also can be considered to act as an important allergen³. We have studied the skin prick test and patch test with cockroach antigen in patients with AD to evaluate the possible role in the pathogenesis of AD and also compared the skin reactions according to 3 different exposure method of patch test on normal appearing skin of AD patients to know what kind of patch test methods could be desirable in APT.

MATERIALS AND METHODS

1. Subjects

Fifty-seven patients with atopic dermatitis were included in our study and all patients fulfilled the diagnostic criteria of atopic dermatitis⁴. There were 26 male patients (45.6%) and 31 female patients (54.4%). The age range of patients was 3 to 36 years (mean; 12.6 years). A complete history was taken and physical examination performed by the same dermatologist. The control group consisted of 30 volunteers with or without allergic rhinitis and asthma, but no history of atopic dermatitis. There were 18 male persons (60%) and 12 female persons (40%) with an age range of 10 to 23 years (mean; 16 years).

2. Cockroach antigens

We collected the American cockroaches (AC) and German cockroaches (GC), and body, feces, and eggs of GC using microscope. The extracts for whole body of AC and GC, and for body, feces, eggs of GC were prepared by petroleum ether using Soxhlet extractor for 4 hours to remove fat as well. Protein concentration was measured with BCA technique⁵. For AC, 53.9 mg/dl and for GC,

17.6 mg/dl of protein was measured. The body, eggs, and feces of GC were measured at 13.7 mg/dl, 11.6 mg/dl, 8.5 mg/dl of protein, respectively.

3. Methods

We studied the severity of atopic dermatitis in 57 patients diagnosed with atopic dermatitis by the grading system of Rajka and Langeland⁶. Total IgE was measured with paper radioimmunoassay (PRIST, Behring, Germany) and specific IgE for cockroach antigen was measured using the Immuno CAP system (Pharmacia & Upjohn, Sweden). The normal range of total IgE was standardized as follows; 1-5 year olds: less than 60 IU/ml, 6-9 year olds: less than 90 IU/ml, 10-15 year olds: less than 200 IU/ml, and in adults: 0-100 IU/ml. The detection limit specific IgE was 0.35KU/l; children were regarded as sensitized if specific IgE level was above detection limit.

Prick test was done in 57 patients with AD and 30 control persons using crude extract of whole body of AC, and the whole body, body, feces, and eggs of GC. A drop of 5 different allergenic extracts was applied to the skin of the volar side of forearm with a lancet. Histamine was used as a positive control, and saline solution was used as a negative control. After 20 minutes the severity of the reaction was evaluated on the basis of erythema and wheal, and graded on a scale of 0 to 4 as follows; 0 = no erythema or wheal, 1 = the size of erythema is smaller than 15 mm, 2 = the size of erythema is larger than 15 mm or the size of wheal is smaller than 3 mm, 3 = the size of wheal is between 3 to 5 mm, 4 = the size of wheal is greater than 5 mm or if pseudopod is developed⁷. Since the score we obtained from the positive control using histamine was 3, we considered the reaction as positive when the score was higher than 3.

In patient group, we applied the antigens to two different areas for the atopic patch test (APT). To the lesional area (mainly antecubital area), we applied the crude extract of the body of GC (Ext) and frozen grinded powder of the whole body of GC (As is) using 8mm Finn chamber (Epitest Ltd Oy, Finland) and Scanpor tape (Alpharma AS, Norway). To the non-lesional area (mainly upper back), we applied the Ext and As is antigen by using the standard method, scratch method, and DMSO mix method. The standard method is simply to attach the antigen to the skin. The scratch method is to

Table 1. Severity according to age distribution in patients with atopic dermatitis

Severity Age	Mild	Moderate	Severe	Total(%)
1-5	1	3	5	9
6-10	3	12	9	24
11-15	0	1	1	2
16-20	1	2	10	13
21-25	0	2	4	6
26-	0	2	1	3
Total(%)	5(8.8)	22(38.6)	30(52.6)	57(100)

attach the antigen on scratched skin with a 27-gauge needle about 4 times in a criss-cross pattern. And the DMSO mix method is to attach antigen after mixing with 20ul of 10% DMSO. In the control group, we used same methods as were applied to the non-lesional area. Patches were removed after 48 hours, and we examined skin reactions according to the criteria of ICDRG⁸ and determined the reaction as positive when the patients score was higher than 1+.

4. Data analysis

Measured data was analyzed with SPSS 10.0 for Windows. The amount of total IgE and specific IgE in serum and the significance of the difference in positive reactions between the patch test and the prick test were tested using the independent sample's t-test. In the patch test, we analyzed the data from each test with χ^2 -test. The data includes the comparison of the positive patch test reaction between the lesional and non-lesional groups, and the difference of three methods used in the non-lesional area, the severity of the disease, and the correlation of positive prick test reactions.

RESULTS

1. Severity of atopic dermatitis

The clinical manifestations in 57 patients with AD were mild in 5 patients (8.8%), moderate in 22 patients (38.6%), and severe in 30 patients (52.6%). The majority of patients developed severe (Table 1).

2. The levels of specific IgE and total IgE in Serum

Among 48 test subjects, 30 of them (62.5%)

Table 2. The positive reaction to cockroach antigen in prick test in 57 patients and 30 control persons

Allergen	No. of patients (%)	No. of control (%)
Whole body of AC	16 (28.1%)	2 (6.7%)
Whole body of GC	16 (28.1%)	3 (10%)
Egg of GC	11 (19.3%)	3 (10%)
Body of GC	11 (19.3%)	3 (10%)
Feces of GC	11 (19.3%)	2 (6.7%)

showed an increase in total IgE level.

Considering the level of specific IgE against cockroach antigen in the test subjects, 38 patients (79.2%) scored class 0, 7 patients (14.6%) class I, 2 patients (4.2 %) class II, and 1 patient (2.1%) class III. Unfortunately, we did not measure the level of specific IgE and total IgE in the control group.

3. Prick test

The positive reaction was seen in 16 patients (28.1%) to whole body of GC and AC, and in 11 patients (19.3%) for each of the eggs, feces, and body of GC. In the control group, 3 patients (10%) showed a positive reaction to each of the whole body, body, eggs of GC and only 2 patients (6.7%) to each of the whole body of AC, feces of GC (Table 2). The positive reaction rate to whole body of AC was significantly higher in the patient group than control group ($p < 0.05$) and the positive rate to whole body of GC was high, but not significantly high in the patient group ($p = 0.053$). There was no significant differences in positive rate between each prick test antigen group ($p > 0.05$).

4. Patch test

In the lesional area, there were positive patch test reactions to Ext and As is in 6 patients (10.5%) and 25 patients (43.9%), respectively. The positive patch test rate to Ext is 0% in standard method, 6.72% in scratch method, 3.3% in DMSO mix method in control group, and 8.8%, 14%, and 12.3% respectively in patient group. The positive patch test rate to As is 13.3% in standard method, 16.7% in scratch method, 13.3% in DMSO mix method in control group, and 49.1%, 50.9%, 50.9% respectively in patient group (Table 3).

There were significant differences in positive patch test reactions to either Ext or As is between

Table 3 The positive reactions to cockroach antigen in patch test

Group	Method Allergen	Lesion	Non-lesion		
		Standard(%)	Standard(%)	Scratch(%)	DMSO(%)
Atopic dermatitis (n=57)	Ext	6 (10.5)	5 (8.8)	8 (14)	7 (12.3)
	As is	25 (43.5)	28 (49.1)	29 (50.9)	29 (50.9)
Control (n=30)	Ext	Not done	0 (0)	2 (6.7)	1 (3.3)
	As is	Not done	4 (13.3)	5 (16.7)	4 (13.3)

Table 4. Comparisons of total IgE and pinprick, patch tests in patients with atopic dermatitis

Total IgE (IU/ml)	Pinprick test (Whole body of GC)		Patch test (Lesional, As is)	
	(+)	(-)	(+)	(-)
	1263.02	549.86	857.18	681.04

patient group and control group ($p < 0.05$). The positive rate to As is was significantly higher than to Ext in either lesional skin or non-lesional skin in patient group ($p < 0.05$). But there were no considerable differences in positive reactions to either Ext or As is between 3 different methods (standard, scratch, DMSO mix) in both lesional and non-lesional areas ($p > 0.05$).

5. The comparison of the prick test, the patch test, and IgE (Table 4)

In 7 patients having class I cockroach antigen specific IgE level, 2 patients showed positive prick test reactions to all kinds of cockroach antigens and one patient showed positive prick test reactions to the whole body of AC and GC. In the skin prick test subjects who scored higher than Class II specific IgE level to cockroach antigen, 2 patients showed a positive reaction to whole body of GC and all 3 patients showed a positive reaction to whole body of AC.

In 7 patients class I cockroach antigen specific IgE level, positive patch test reactions were observed in 2 patients (Ext) and 4 patients (As is). But there was only one patient showing positive patch test reaction to cockroach antigens among 3 patients who had more than class II specific IgE level to cockroach antigen.

The total IgE level of As is negative patch test group in lesional area were 681.04 IU/ml and this amount was lower than As is positive group which

were 857.18 IU/ml. But there were no significant differences between them ($p > 0.05$). In the GC (whole body) prick test positive group, there was 1263.02 IU/ml of total IgE, and this amount was significantly higher than GC (whole body) prick test negative group who had 549.46 IU/ml ($p < 0.05$) (Table 4).

There was only 1 patient showing positive prick test reaction to cockroach antigens among the 6 patients who showed positive patch test reactions to Ext antigen in lesional area of atopic dermatitis. There were 9 patients showing positive prick test reactions to one or more cockroach antigens among 25 patients who showed positive patch test reactions to As is antigen. There were no significant relationships between the positive reactions to patch test and prick test to Ext and As is antigen.

DISCUSSION

There are 4 kinds of household cockroaches in Korea including German cockroaches (*Blattella germanica*), American cockroaches (*Periplaneta americana*), smoky brown cockroaches (*Periplaneta fuliginosa*), and Japanese cockroaches (*Periplaneta japonica*). Among them, the German cockroach is the most common species in Korea⁹. Whole body cockroach extracts have been shown to cause cutaneous sensitivity^{10, 11} as well as inhaled allergy¹², but potential sources of relevant cockroach allergens in the environment include whole bodies, cast skins, secretions, egg castings, and fecal materials.

Table 5. Comparison of RAST, prick test and patch test in 57 patients

No	RAST	PRICK					PATCH	
		American	German	body	egg	feces	extract	As is
1	0	1	1	0	0	0	0	1
2	0	0	0	0	0	0	0	1
3	0	1	1	1	1	1	0	0
4	0	0	0	0	0	0	1	2
5	0	0	0	0	0	0	1	2
6	1	0	0	0	0	0	1	2
7	0	0	0	0	0	0	0	1
8	0	0	0	0	0	0	0	1
9	0	0	0	0	0	0	0	1
10	0	0	0	0	0	0	0	0
11	0	0	0	0	0	0	0	1
12	1	0	0	0	0	0	0	0
13	0	0	0	0	0	0	0	0
14	0	0	0	0	0	0	0	0
15	0	0	0	0	0	0	0	0
16	0	0	0	0	0	0	0	0
17	0	0	0	0	0	0	0	0
18	0	0	0	0	0	0	0	0
19	0	0	0	0	0	0	0	0
20	0	0	0	0	0	0	0	1
21	0	0	0	0	0	0	0	0
22	1	0	0	0	0	0	0	0
23	0	0	0	0	0	0	0	1
24	0	0	0	0	0	0	0	1
25	0	0	0	0	0	0	0	0
26	ND	0	0	0	0	0	0	0
27	2	1	1	1	1	1	1	1
28	ND	0	0	0	0	0	1	1
29	ND	0	0	0	0	0	0	1
30	ND	0	0	0	0	0	0	0
31	1	0	0	0	0	0	1	1
32	0	0	0	0	0	0	0	0
33	0	0	0	0	0	0	0	1
34	1	1	1	1	1	1	0	1
35	3	1	1	1	1	1		0
36	0	0	1	0	0	0	0	1
37	0	0	1	0	0	0	0	0
38	ND	1	1	1	1	1	0	1
39	0	0	0	0	0	0	0	0
40	ND	0	0	0	0	0	0	0
41	1	1	1	0	0	0	0	1
42	0	0	0	0	0	0	0	0
43	0	0	0	0	0	0	0	0
44	0	1	0	0	0	0	0	0
45	0	0	0	0	0	0	0	1
46	1	1	1	1	1	1	0	0
47	0	0	0	0	0	0	0	0

No	RAST	PRICK					PATCH	
		American	German	Body	Egg	Feces	Extract	As is
47	0	0	0	0	0	0	0	0
48	2	1	0	0	0	0	0	0
49	ND	0	0	0	0	0	0	0
50	0	1	1	0	0	0	0	0
51	ND	0	0	0	0	0	0	0
52	0	1	1	1	1	0	0	1
23	0	1	1	1	1	1	0	1
54	0	1	1	1	1	1	0	1
55	0	1	1		1	1	0	0
56	0	1	1	1	1	1	0	0
57	ND	0	0	0	0	0	0	0

ND : not done

Atopic individuals who live in cockroach-infested housing become sensitized by inhalation of potent cockroach allergens and produce vigorous IgE antibody responses¹³. Kang et al¹⁴ also reported 20% of positive reactions against cockroaches. Next to the house dust mite (e.g. *D. farinae*, *D. pteronyssinus*) and house dust, cockroaches ranked fourth, showing the possibility of cockroaches as a major indoor aeroallergen. Park et al¹⁵ confirmed several common German cockroach allergens with molecular weights of 67, 64, 55 and 30 kd in Korean atopy. Among them, particularly a 55 kd allergen can be considered to be the major allergen.

Although asthma and allergic rhinitis is distinctly related to the inhalation of allergens, there is still debate over the role of IgE and inhaled allergens in the pathogenesis of AD¹⁶. Patients with atopic eczema often have elevated serum levels of IgE, which may correlate with severity of disease. In addition, aeroallergen avoidance, especially with regard to house dust mites, can result in marked improvement of skin lesions¹⁷⁻²¹.

In our study, total IgE level was increased in 30 patients (62.5%). There were 10 patients (20.8%) showing German cockroach specific IgE in serum, but 3 patients showed specific IgE level more than class II (6.3%). The positive prick test reaction to whole body of GC and AC was seen in 16 patients (28.1%) but in 11 patients (19.3%) to egg, body, or feces of GC, and whole body extract of AC and/or GC is preferable as a skin prick test antigen. Musmand et al²² demonstrated that cockroach feces have significant allergenic activity and similar allergen content to cockroach whole body and this re-

sult coincides with our previous study²³. Additionally in prick test positive group of the whole body of GC, there was significantly high total IgE than prick test negative group of the whole body of GC, but regarding the correlation of APT with skin prick test and specific IgE results, no significant concordance was observed.

The mechanism by which aeroallergens can induce eczematous skin lesions may involve aeroallergens, penetration through the epidermis, and binding to IgE on Langerhans cells. These IgE-bearing Langerhans cells then present allergens to T cells, which induce a delayed inflammatory response. The cytokines released by these activated T cells may also play a role in the induction and/or regulation of IgE production by B cells in the afferent lymph nodes²⁴.

An epicutaneous patch test with aeroallergens especially in patients with atopic eczema was first documented in 1982 by Mitchell et al²⁵. This epicutaneous patch test with aeroallergens was reproduced and extended by several groups²⁶⁻²⁹, and the term 'atopy patch test' (APT) was introduced³⁰. Most studies show that nonatopic patients are not reactive to APT. Among the patients with AD, 10 to 100% of them show positive reactions to APT³¹. The most likely reasons for this wide variation in positive reactions was not the selection of patients with AD but the methodology of the APT. Variables in these studies that might have affected the outcome of the APT were allergen concentrations, tape stripping, reading time, and application site. Our study was to reevaluate the effects of some variables in the methodology of APT¹⁶, to provide an

optimal method for the APT. To facilitate allergen penetration, we applied scratch method and DMSO mix method. Dimethyl sulfoxide (DMSO) is used therapeutically as a local analgesic, an anti-inflammatory agent, and a promotor for percutaneous penetration of certain chemicals. Real contact urticarial lesions are frequently caused by DMSO at higher concentration³² and 20% DMSO caused transient erythema at the site of application in most subjects³³. So we applied lower concentration 10% DMSO in DMSO mix method to minimize the non-immunologic reaction. The result was almost similar between the standard, scratch, and DMSO mix method. It could be concluded that standard patch test procedure is preferred for the future patch testing.

There were significant differences in positive reaction to As is antigens in all 3 patch test methods between patients group and control group, and no considerable differences to Ext antigen except standard patch test method. Our results suggest that As is antigen concentration may be too high or irritating in atopic patients having abnormal barrier function and standard patch test with Ext antigen could be more preferable.

In conclusion, cockroach antigens also could be considered to be involved in atopic dermatitis, and APT with cockroach antigen may provide further diagnostic information in addition to a patient's history, skin prick test, and RAST results. With a standardized APT, the actual clinical relevance of IgE-mediated sensitizations for the eczematous skin lesions might be better evaluated. In the future, we hope APT could be used as the critical provocation test for atopic dermatitis like the bronchial provocation and nasal provocation are the tests of choice in atopic respiratory diseases.

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